Diurnal Variations of Post-exercise Parasympathetic Nervous Reactivation in Different Chronotypes

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SUMMARY

The purpose of this study was to investigate the diurnal variation and chronotype differences, i.e., in morning-types and evening-types, in post-exercise vagal reactivation. Twelve healthy male college students who were classified as morning-type (6) and evening-type (6), based on responses to a questionnaire, participated in this study. Post-exercise vagal reactivation was assessed as the time constant of the beat-by-beat heart rate decrease for the first 30 sec after exercise (T_{30}) at an intensity lower than the ventilatory threshold. The subjects performed 3-min cycle ergometer exercise at an intensity corresponding to 80 % of the ventilatory threshold after a 1 min warm-up exercise in the morning (7:00 - 8:00) and evening (17:00 - 18:00) to obtain the T_{30}. A significant interaction (chronotype-by-time) effect was found for T_{30}. The morning value of the T_{30} in evening-type subjects was significantly larger than their evening value and the morning value in morning-type subjects. There was no significant interaction effect for heart rate and oxygen uptake during exercise. These results suggest that diurnal variation in post-exercise vagal reactivation is different between morning-type and evening-type, and post-exercise vagal reactivation in evening-type individuals is sluggish in the morning. (Jpn Heart J 2001; 42: 163-171)

Key words: Autonomic nervous activity, Biological rhythm, Exercise

INCREASED heart rate (HR) during exercise is decreased to its resting value after cessation of the exercise. In the early phase of recovery, the prompt restoration of vagal tone which is depressed during exercise mainly adjusts HR.[1)] The dynamic vagal reactivation immediately after exercise, assessed by the time constant of the HR decrease for the first 30 sec after exercise at an intensity lower than the ventilatory threshold, was accelerated in well-trained athletes but slowed in patients with chronic heart failure, in comparison to age-mached normal volunteers.[2)] The dynamic vagal reactivation immediately after exercise may be an

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important mechanism in preventing excessive cardiac work after exercise. When the HR recovery is slow, myocardial oxygen consumption after exercise increases and potentially exerts a harmful effect on the failing or ischemic heart. Because ventricular arrhythmia appears more commonly in the post-exercise period than the actual exercise period,[3] post-exercise vagal reactivation may play an important role in suppressing ventricular arrhythmias related to exercise.

There are significant circadian variations in physiological responses to exercise. Intense exercise provides a greater shock to the cardiovascular system at night and early morning than at other times,[4] and exercise in the early morning induced coronary arterial spasm in patients with Prinzmetal's variant angina.[5] There is also a prominent circadian variation in the time at which cardiac death occurs, with a low incidence during the night and an increased incidence in the morning.[6] The circadian variations in the cardiovascular responses to exercise and the incidence of cardiac attack may be partly linked with the circadian variation in autonomic nervous system activity and response to exercise. Therefore, it is important to investigate the circadian variation in autonomic nervous system activity and response to exercise.

There are individual differences in circadian characteristics. Circadian variation in body temperature was found to be different between morning-type and evening-type chronotypes classified according to the responses to questions regarding sleep and working times and the phasing of work and habitual activity.[7] Maximal oxygen uptake in the evening-types was best in the evening, whereas morning-types were not affected by time of day.[8] Physiological responses to submaximal exercise, e. g. HR and $\dot{V}O_2$, were not affected by individual chronotype.[8] However, no study has investigated chronotype differences in autonomic nervous responses to exercise.

The purpose of this study was to investigate the diurnal variation and chronotype differences in vagally mediated HR recovery after exercise. We hypothesized that post-exercise HR recovery is slowed in the morning, especially in evening-types.

Subjects and Methods

Subjects: Thirty-seven healthy college students (male, aged 20 - 28 yr) were classified as morning-types ($n = 6$) or evening-types ($n = 8$) based on their responses to the questionnaire devised by Horne and Ostberg,[7] which is the main method of chronotype classification. The remaining 23 individuals were classified as neither type.

Six morning-type individuals and 6 of 8 evening-type individuals who gave written informed consent to participate in this study underwent exercise tests.

Each chronotype group included one athlete (morning-type: canoe; evening-type: tennis), and the remaining subjects were non-athletes at the recreational sports level. The athletes continued their usual training during the experimental period, but did not perform any heavy training or sports activities in the weeks preceding the experiments. The non-athletes generally performed light exercise, e.g. jogging, and also did not participate in any heavy sports activities in the weeks preceding the experiments. Each subject usually woke up by 8:00 a.m. in the weeks preceding the experiments because all measurements were performed during the college trimester. No subject was taking any pharmacological agent.

Experiment Procedures: The subjects abstained from doing vigorous physical activity and drinking alcoholic beverages on the day before and the day of the experiment, drinking coffee and smoking on the day of the experiment, and eating any food for 3 h before the experiment.

First, each subject performed an incremental cycle ergometer exercise test between 15:00 and 18:00 in order to measure maximal oxygen consumption ($\dot{V}O_2max$) and ventilatory threshold (VT). The incremental exercise test began at a workload of 50 W, and the workload was then increased by 20 W every 1 min until volitional exhaustion. Gas exchange data were collected throughout the exercise test using a breath-by-breath respiromonitor system (AE 280S Aeromonitor, Minato Medical Science Co., Tokyo, Japan). Oxygen uptake ($\dot{V}O_2$), carbon dioxide output ($\dot{V}CO_2$) and minute ventilation ($\dot{V}E$) were obtained during exercise. The $\dot{V}O_2max$ was defined as the highest $\dot{V}O_2$ achieved during the test. The VT was determined by gas exchange data from the V-slope method.[9)]

Second, each subject performed constant load cycle ergometer exercise tests to evaluate the post-exercise vagal reactivation one day between 1 and 2 weeks after the incremental exercise test. The tests were carried out in the morning (7:00 - 8:00) and in the evening (17:00 - 18:00). The subjects got up at least 30 min before the morning test and avoided eating. Before the constant load exercise tests, each subject rested quietly for 20 min in the supine position, and oral temperature and R-R interval (sampling frequency: 1 msec) were measured using a HR monitor (AM01-M01, NADEX Co., Tokyo) during the last 5 min of the supine rest under breathing control (15 breaths / min). Then, after 2 min of resting in the sitting position on a cycle ergometer, the subjects performed consecutively a 1 min warm-up (40 % VT) and a 3 min main-exercise (80 % VT). The R-R intervals were recorded during sitting rest and from the start of rest to 2 min after cessation of the exercise. The mean HR during exercise was calculated from the 1 min R-R interval data before the end of the exercise. The R-R interval data for the first 30 sec after exercise was used to analyze the index of post-exercise vagal reactivation. Gas exchange data were recorded using a respiromonitor, and VO_2 for the last 1 min of exercise was evaluated.

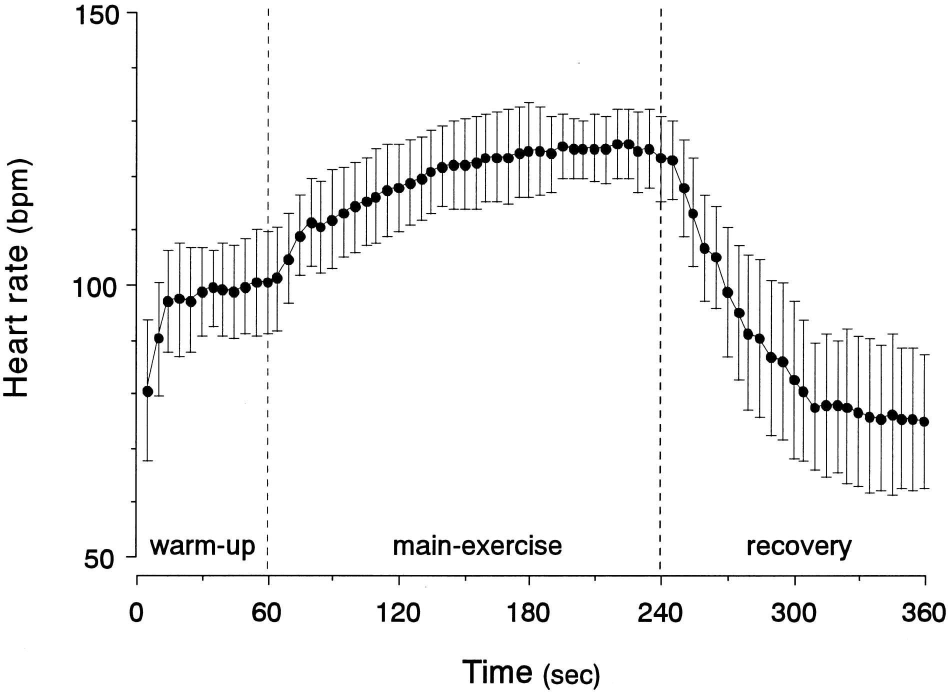

Figure 1. The mean time course of heart rate during the constant load exercise test. Data are expressed as mean ± SD.

Power spectral analysis of HR variability : The R-R interval data during the last 5 min of supine rest were examined using power spectral analysis (Memcalc method; GMS Co., Tokyo) to obtain the index of cardiac parasympathetic nervous activity, i.e. the natural logarithm (ln) of high frequency power (HF: 0.15 - 0.50 Hz).

Analysis of HR decline: The time constant of beat-by-beat HR decrease for the first 30 sec after the 80 % VT exercise (T_{30}) was obtained as an index of vagal reactivation after exercise, according to the method of Imai, *et al.*[2] The mean time course of the momentary HR calculated from the R-R interval data during exercise tests is shown in Figure 1. The HR increased progressively from the start of exercise and reached a steady state during exercise. No subject exhibited any arrythmia during the experiment. After the cessation of exercise, the HR appeared to decrease exponentially. Linear regression analysis was performed on the natural logarithm of HR data for the first 30 sec after exercise, and then the negative reciprocal of the slope of the regression line was determined as the T_{30}.

Statistical analysis: All data are expressed as the mean and standard deviation. Repeated measures ANOVA was used in the data analysis. When there was a sig-

nificant interaction (chronotype-by-time) effect, contrast comparison tests were used to determine differences among means. Linear regression analysis was performed by the least-squares method. Significance was established at p values < 0.05.

RESULTS

The physical characteristics of the subjects are shown in Table I. There were no significant differences in any of the variables examined between the morning-types and evening-types.

The T_{30} values for both chronotypes are shown in Figure 2. A significant

Table I. Physical Characteristics of Subjects

Variable	(unit)	Morning type (n = 6)	Evening type (n = 6)
Age	(yr)	24.0 ± 1.7	24.0 ± 3.3
Height	(cm)	173.8 ± 8.0	171.8 ± 6.2
Weight	(kg)	70.0 ± 12.2	67.9 ± 7.8
$\dot{V}O_2max$	($ml \cdot min^{-1}$)	3255.0 ± 471.8	3085.5 ± 619.9
	($ml \cdot kg^{-1} \cdot min^{-1}$)	46.9 ± 4.5	45.5 ± 8.3
$\dot{V}O_2VT$	($ml \cdot min^{-1}$)	2088.3 ± 337.9	1956.8 ± 221.0
	($ml \cdot kg^{-1} \cdot min^{-1}$)	30.0 ± 2.3	28.9 ± 2.1
Work load VT	(W)	175.0 ± 17.0	170.0 ± 26.0

Mean ± SD. $\dot{V}O_2max$ = maximal oxygen uptake; $\dot{V}O_2VT$ = oxgen uptake at ventilatory threshold; Work load VT = Work load at ventilatory threshold.

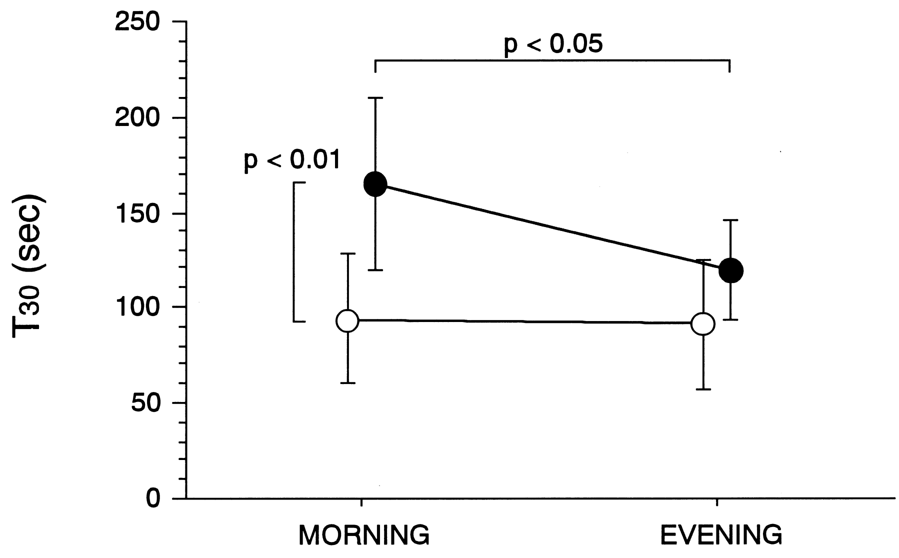

Figure 2. The time constant of beat-by-beat heart rate decrease for the first 30 sec after exercise (T_{30}). ○: morning-type, ●: evening-type. Data are expressed as mean ± SD.

Table 2. Descriptive Data for Dependent Variables in AM and PM and Summary of ANOVA Results

Variable		AM	PM	ANOVA Results Time of Day	ANOVA Results Chronotype
Oral temperature (°C)	All subjects	36.0 ± 0.4	36.7 ± 0.2	$p < 0.001$	NS
	Morning-types	36.0 ± 0.4	36.7 ± 0.3		
	Evening-Types	36.0 ± 0.5	36.7 ± 0.2		
Resting In HF (ln [msec^2])	All subjects	6.4 ± 0.8	6.53 ± 1.2	NS	NS
	Morning-types	6.2 ± 1.0	6.2 ± 1.2		
	Evening-Types	6.6 ± 0.6	6.9 ± 1.2		
Resting HR (bpm)	All subjects	70.6 ± 13.2	72.8 ± 12.2	NS	NS
	Morning-types	67.0 ± 13.4	69.1 ± 11.0		
	Evening-types	74.3 ± 13.1	76.6 ± 13.2		
Exercise HR (bpm)	All subjects	127.1 ± 10.2	126.5 ± 9.3	NS	NS
	Morning-types	122.1± 3.5	122.6 ± 4.0		
	Evening-types	132.0 ± 12.5	130.3 ± 11.9		
$\dot{V}O_2$ (l•.min^{-1})	All subjects	1.71 ± 0.32	1.69 ± 0.34	NS	NS
	Morning-types	1.68 ± 0.35	1.68 ± 0.39		
	Evening-types	1.74 ± 0.31	1.70 ± 0.32		

Mean ± SD. ln = natural logarithm; HF = high frequency power of heart rate variability, HR = heart rate; $\dot{V}O_2$ = oxygen uptake.

interaction (chronotype-by-time) effect was found for T_{30} (F = 5.00, $p < 0.05$). The T_{30} in evening-types was significantly larger in the morning than in the evening (165.5 ± 45.2 sec vs 119.5 ± 25.7, mean ± SD, F = 12.05, $p < 0.05$), while that of morning-types showed similar values in the morning and evening (94.4 ± 33.8 vs 91.2 ± 33.8 sec). In the morning, the T_{30} of evening-types was significantly larger than that of morning-types (165.5 ± 45.2 sec vs 94.4 ± 33.8, F = 5.06, $p < 0.01$).

The descriptive data (resting oral temperature, resting lnHF, resting HR, exercise HR and exercise $\dot{V}O_2$) and the results of the analysis of variance are presented in Table II. A significant time of day effect was found for oral temperature, which was higher in the evening (F = 29.707, $p < 0.001$), however, a significant interaction effect was not obserred. There was neither a significant time of day nor interaction effect for the resting lnHF, resting HR, exercise HR and exercise $\dot{V}O_2$.

DISCUSSION

The diurnal variation in the time constant of post-exercise HR decrease for the first 30 sec (T_{30}) was investigated, in association with chronotypes. A significant interaction (chronotype-by-time) effect was found for T_{30}. Evening-type subjects showed a significantly larger T_{30} after the 80 % VT exercise in the morning than in the evening, while morning-type subjects did not. The morning value of the T_{30} in evening-type was significantly larger than that in morning-type sub-

jects.

The larger morning T_{30} in evening-types may be mainly caused by slowed vagal reactivation after exercise. After exercise, increased HR rapidly decreases due to rapid vagal reactivation and gradual sympathetic withdrawal. Imai, *et al.*[2] showed that the T_{30} was prolonged by vagal blockade but was almost independent of sympathetic blockade, when the exercise intensity was lower than VT, suggesting that the early phase of HR recovery after exercise at a low intensity was mediated primarily by vagal reactivation, and that the T_{30} could be a specific index for post-exercise vagal reactivation. Thus, the results of the present study suggest that post-exercise vagal reactivation is sluggish in the morning in evening-type subjects, but not in morning-type subjects.

We imposed the same work intensity both in the morning and evening tests on the subjects. Therefore, the work intensity determined in the evening might be higher for the morning potential of evening-types. Extremely activated sympathetic activity and accumulation of anaerobic metabolites during intense exercise could attenuate post-exercise vagal reactivation, because the T_{30} was slightly but significantly prolonged by higher exercise intensity over the VT level.[2] In this study, experiments with sympathetic blockade were not performed, so we could not exclude the effect of activated sympathetic activity on T_{30}. However, the prolongation of T_{30} in the morning in evening-types is not likely due to increased sympathetic activity because the exercise in this study was performed at a low level. The T_{30} is nearly independent of exercise intensity when that level is lower than the VT level,[2] and further, the work intensity and $\dot{V}O_2$ at VT is not different between in the morning and evening, in either morning-types or evening-types.[8] Thus, a possible enfeebled effect induced by a higher work load on the HR recovery in evening-types could be eliminated. Furthermore, factors that might have affected performance, such as muscular problems or depletion of muscular glycogen reserves could be eliminated because the subjects abstained from doing vigorous physical exercise starting the day before the exercise test.

In this study, the subjects were not homogeneous with respect to training level since each chronotype group contained one athlete, although the remaining subjects were non-athletes at the recreational sports level. This heterogeneity could influence the value of the T_{30} in each chronotype group, because the T_{30} is short in well-trained athletes,[2] and there is a significant inverse relationship between the T_{30} and $\dot{V}O_2max$.[10] Between the morning-types and evening-types, however, there were no differences in $\dot{V}O_2max$, VT, evening T_{30}, or composition of each group with respect to training level. Therefore, we did not exclude the athletes from this study.

In vagal reactivation after exercise, central mechanisms play a major role, rather than arterial baroreflex or exercise reflexes originating from mechanore-

ceptors or chemoreceptors in exercise muscles, because the T_{30} depended minimally on the exercise intensity, systolic blood pressure, and $\dot{V}O_2$ at the end of exercise.[2] The prolonged T_{30}, i.e. the slowed vagal reactivation, in the morning may be associated with changes in a central mechanism, such as a decrease in the release of inhibitory commands from the cortex to the parasympathetic center and/or in the activity of the parasympathetic center per se. An investigation performed in this laboratory showed that the index of resting vagal activity (resting lnHF) was significantly correlated with the T_{30} (unpublished data). In this study, however, there was no significant difference between the resting lnHF in the morning and evening, regardless of chronotype.

The data obtained in this study cannot identify the reason why the morning T_{30} in evening-types was slower than that in morning-types. Many biological rhythms are a mixture of endogenous components (biological clock) and exogenous factors, e.g. rhythmic environment and life style.[11] The sleep-wake schedule in each chronotype may affect the diurnal or circadian rhythm of post-exercise vagal reactivation. For some subjects, especially the evening-type subjects, the wake-up time was rather early with respect to their habits, and consequently, the earlier wake-up time might have affected the T_{30}. However, the wake-up time on the day of the morning test was probably not markedly earlier than usual for the subjects, because all measurements were performed during a college trimester, and, moreover, all subjects avoided eating before the morning test. A recent study[12] investigated a single nucleotide polymorphism located in the 3' flanking region of the human CLOCK gene as a predictor of diurnal preference (morningness or eveningness), and showed that morningness - eveningness preferences were correlated with the circadian gene polymorphism. Thus, genetic factors might affect the diurnal or circadian variation in post-exercise vagal reactivation.

The time immediately after exercise is considered to be a period when sudden death frequently occurs.[13] Although the reason for the finding is unclear, it may be partly due to a primary arrhythmic event which occurs in the post-exercise period, because ventricular arrhythmia appears more commonly in the recovery period than the actual exercise period.[3] The slowed vagal reactivation might increase the incidence of post-exercise fatal ventricular arrhythmias. It is unclear, however, whether or not the sluggish post-exercise vagal reactivation of evening-types in the morning increases the incidence of heart attacks. In this study, no one showed any arrythmia during the experiment.

Imai, *et al.* [2] demonstrated that vagal mediated HR recovery after exercise was accelerated in well-trained athletes, and suggested that it is a physiologic adaptation allowing for rapid HR recovery after intense exercise. A study by our laboratory [10] showed that vagal mediated HR recovery was significantly correlated with $\dot{V}O_2max$, and that it was slowed after hard training in athletes. The cir-

cadian variation in autonomic nervous activity in association with conditioning as well as with the safety of morning exercise in evening-types should be investigated more intensively.

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