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Effects of post-absorptive and postprandial exercise on 24 h fat oxidation

Kenshiro Shimada [a], Yuki Yamamoto [a], Kaito Iwayama [a], Kazuteru Nakamura [a], Sachiko Yamaguchi [b], Masanobu Hibi [b], Yoshiharu Nabekura [a], Kumpei Tokuyama [a,*]

[a] Doctoral Program in Sports Medicine, Graduate School of Comprehensive Human Sciences, University of Tsukuba, Tennodai 1-1-1, Tsukuba, Ibaraki, Japan 305-8574

[b] Health Care Food Research Laboratories, Kao Corporation, Bunka 2-1-3, Sumida, Tokyo, Japan 131-8501

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ABSTRACT

Objective. Fat oxidation during exercise depends on nutritional state, and exercise performed in the post-absorptive state oxidizes more fat than that performed in the postprandial state. However, the effects of exercise on energy metabolism continue during the post-exercise period, and the difference in fat oxidation during exercise may be compensated for during the post-exercise period. The present study compared the effects of an acute exercise bout in the post-absorptive or postprandial state on 24 h fat oxidation.

Methods. Twelve young male athletes stayed twice in a room-size metabolic chamber for 24 h indirect calorimetry in a randomized repeated-measure design. Before or after breakfast, i.e. in the post-absorptive or postprandial state, subjects exercised at 50% VO_2 max for 60 min.

Results. During the 60 min of exercise, energy expenditure in the two exercise trials were equivalent, but exercise in the post-absorptive state was performed with lower RQ compared with that in the postprandial state ($P<0.01$). The time of exercise relative to breakfast did not affect 24 h energy expenditure ($P>0.5$). However, accumulated 24 h fat oxidation was higher ($P<0.05$) and that of carbohydrate oxidation was lower ($P<0.05$) when exercise was performed in the post-absorptive state.

Conclusions. Compared with exercise performed in the postprandial state, exercise performed in the post-absorptive state oxidized more fat and saved more carbohydrate in the body, without affecting 24 h energy expenditure.

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1. Introduction

Fat oxidation during exercise is suppressed by carbohydrate ingestion before exercise [1–7]. This effect persists at least for 4 h after a meal, based on studies comparing metabolic response to exercise performed 2, 4, 6, 8 or 12 h after carbohydrate ingestion [6]. For individuals eating 3 meals a day, metabolic response to exercise is often under the influence of a recent meal, and exercising in the fasted state is usually only possible before breakfast. These findings suggest that exercise before breakfast, a common practice among athletes and recreational runners, is beneficial to reduce body fat. Observations in aforementioned studies were limited on energy metabolism only during the exercise. However, an impact of

Abbreviations: RQ, respiratory quotient; EPOC, excess post-exercise oxygen consumption; VO_2, oxygen consumption rate; VO_2, max (maximal oxygen uptake); VCO_2, carbon dioxide production rate; N, urinary nitrogen excretion rate; MET, metabolic equivalent; W, watt.

* *Corresponding author*. Tel.: +81 298 53 3963; fax: +81 298 53 6507.
E-mail address: tokuyama@taiiku.tsukuba.ac.jp (K. Tokuyama).

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exercise on energy metabolism is not confined to the period of physical activity itself, and oxygen consumption remains above the resting metabolic rate during post-exercise periods, known as excess post-exercise oxygen consumption (EPOC) [8,9]. Furthermore, a relative shift from carbohydrate to fat as a substrate source is a consistent finding during the post-exercise period [2,10]. It is therefore necessary to take the post-exercise period into account, when the effect of exercise on body fat balance is concerned. A few studies, which compared metabolic effects of exercise performed in post-absorptive and postprandial state, extended their observation into the post-exercise period. Compared with exercise performed in postprandial state, exercise performed in post-absorptive state oxidized more [11] or less fat [1,12] during 2–6 h of the post-exercise period. Furthermore, experimental protocol in these studies made interpretation of the results not straightforward. First, when the order of exercise and meal was reversed, the two exercise trials were not matched for duration of post-meal and post-exercise period. Second, indirect calorimetry was terminated before energy metabolism of the two exercise conditions became indistinguishable, suggesting that EPOC and/or thermic effect of food was still ongoing at the end of experiment. Thus, it is still inconclusive whether exercise performed in post-absorptive state increases 24 h fat oxidation more than that performed in postprandial state.

The primary aim of our study was to determine whether 24 h fat oxidation was affected by time of exercise relative to breakfast, i.e. in post-absorptive and postprandial state. Using a room-size metabolic chamber, effects of exercise performed before and after breakfast on 24 h energy metabolism were compared. Indirect calorimetry with a room-size metabolic chamber provides a controlled environment, in which the impact of exercising protocol on energy metabolism can be continuously measured for a long period under identical energy intake [13].

2. Methods

2.1. Subjects characteristic

Twelve young male endurance athletes were recruited in this study after giving their written informed consent. This study was approved by the ethics committee of University of Tsukuba. Physical characteristics of subjects were 22.8 ± 0.6 years of age, 170.2 ± 1.5 cm of height, 60.9 ± 1.5 kg of body weight and 13.1% ± 0.7% of body fat. Their maximal oxygen uptake (VO_2 max) was 64.1 ± 1.7 mL/kg/min. All subjects were free from pathological condition and none of them were taking any medications or supplements. One week before the 24 h calorimetry, subjects spent one night in a metabolic chamber in order to get acclimated to the measurement condition.

2.2. Pre-study evaluation

The workload corresponding to 50% of the individual maximal VO_2 was determined from an incremental exercise test on a bicycle ergometer (Aero bike 75XLIII, Combi, Tokyo, Japan), 1 or 2 weeks before the experiment. All subjects performed a graded exercise test which was comprised of submaximal and maximal tests. Initial workload of the submaximal test was 60 W and it was increased by 20 W every 4 min until ventilatory threshold was observed, ratings of Borg scale of perceived exertion reached 17 (very hard) or the workload reached 200 W. After a 5 min rest, subjects performed a maximal test that started from the last workload of the submaximal test, and it was increased by 10 W every min until exhaustion. The highest VO_2 for consecutive 60 s during the maximal test was taken as subject's VO_2 max. Respiratory gas analysis was continuously performed on a breath-by-breath basis using the computerized standard open circuit technique (Oxycon Alpha, Jaeger, Wuerzberg, Germany).

2.3. Experimental protocol

The subjects stayed twice in a room-size respiratory chamber in a randomized repeated-measure design, and the two trials were separated at least 1 week. Between the two trials, there was no significant difference in body weight and body composition.

Subjects entered the chamber the day before exercise session (day 1, 2200 h), and exited the chamber the day after exercise session (day 3, 0630 h). On day 2, breakfast (at 0800 h), lunch (at 1200 h) and supper (at 1900 h) were provided. Exercise was performed before (0630–0730 h) or after (1030–1130 h) breakfast at 50% of VO_2 max for 60 min using a bicycle ergometer. Subjects remained sedentary during the rest of the day, except for 30 min of shower break (1815–1845 h). Energy metabolism during the 30 min break was estimated from metabolic equivalent of taking shower as 2.0 METs [14] and respiratory quotient (RQ) immediately before the break. Twenty four hour energy expenditure and nutrients oxidation were calculated from 0600 h of day 2 to 0600 h of day 3.

Six meals prior to and during the 24 h indirect calorimetry were individually standardized according to the estimated energy requirement for Japanese [15], assuming physical activity factor to be 1.68 (2458 ± 56 kcal/day). Expressed as a percentage of total energy, the standardized meals contained 15% protein, 25% fat and 60% carbohydrate. The contribution of the breakfast to total 24 h energy intake was 31%. From one day prior to the 24 h indirect calorimetry, the subjects were instructed to refrain from consumption of beverages containing energy, caffeine or alcohol. On the day before the calorimetry, the subjects were allowed to exercise, but were requested to repeat the same mode, duration and intensity on the day before the second visit to the laboratory. Exercise had to be completed at least 12 h prior to entry to the metabolic chamber.

2.4. Measurements

Energy metabolism was measured with a room-size metabolic chamber (Fuji Medical Science, Chiba, Japan). The airtight chamber measures 2.00 × 3.45 × 2.10 m, having an internal volume of 14.49 m^3. The chamber is furnished with an adjustable hospital bed, desk, chair, toilet and bicycle ergometer. Air in the chamber was pumped out at a rate of

100 L/min. Temperature and relative humidity of in-coming fresh air were controlled at 25.0±0.5 °C and 55.0%±3.0%, respectively. Concentrations of oxygen (O_2) and carbon dioxide (CO_2) in out-going air were measured by an online process mass spectrometer (VG Prima δB, Thermo Electron, Winsford, UK). Precisions of the mass spectrometry, defined as the standard deviation for continuous measurement of calibration gas mixture (O_2 15%, CO_2 5%), were 0.0016% and 0.0011% for O_2 and CO_2, respectively. At every 5 min, O_2 consumption (VO_2) and CO_2 production (VCO_2) rates were calculated using an algorithm for improved transient response [16].

Macronutrients oxidation and energy expenditure were calculated from VO_2, VCO_2 and urinary nitrogen excretion [17]. Rates of glucose, lipid and protein oxidation were computed from the following equations, assuming urinary nitrogen excretion rate (N) to be constant during the calorimetry.

Glucose oxidation (g/min) = 4.55 VCO_2 (L/min) – 3.21VO_2 (L/min) – 2.87 N (g/min).

Fat oxidation (g/min) = 1.67VO_2 (L/min) – 1.67VCO_2 (L/min) – 1.92 N (g/min).

Protein oxidation (g/min) = 6.25 N (g/min).

Subsequently, energy production by each nutrient was calculated by taking into account the caloric equivalent of the three substrates, i.e., 3.74 kcal/g glucose, 9.50 kcal/g fat and 4.10 kcal/g protein. It is conceivable that tissue glycogen rather than glucose is the predominant form of carbohydrate being oxidized during exercise. An equation for complete oxidation of glycogen and its caloric equivalent are different from those of glucose. However, estimates of energy expenditure by carbohydrate oxidation in terms of energy expenditure in a unit time (kcal/min) are robust, regardless of whether glucose or glycogen is oxidized in the body [17,18]. Therefore, glucose oxidation was discussed as carbohydrate oxidation in the present study.

Apparent energy balance and nutrient balance were estimated as difference between the input and output. For example, fat balance was defined as

fat balance = total metabolizable fat intake – whole body fat oxidation.

This represents "apparent" fat balance, since not all the dietary fat is absorbed during the short period [19,20].

Physical activities including non-exercise activity were estimated using a wrist watch-like device, ActiGraph (Ambulatory Monitoring, NY, USA) with zero crossing mode, as the number of times the activity signal crossed the zero reference point per minute (counts/min).

2.5. Statistical analysis

Data in the text and figures were given as means ± SE of the experimental condition. Paired t-test was used to compare mean values of two experimental conditions.

To compare time course of energy metabolism in the two experimental conditions, mean values in the morning (0600 h–1200 h), afternoon (1200 h–1900 h), evening (1900 h–2300 h) and sleep (2300 h–0600 h) were calculated for each subject, and repeated measures two-way analysis of variance (ANOVA) was used. Statistical analysis was performed using SPSS statistical software (Version 14.0, SPSS Japan, Tokyo, Japan), with the level of statistical significance set at 5%.

3. Results

During the 60 min of exercise, energy expenditure (555 ± 19 and 563 ± 17 kcal for exercise before and after breakfast trials, $P > 0.5$), mean heart rate (120 ± 4 vs. 122 ± 4 beats/min, $P > 0.5$) and relative intensity of exercise (48.8% ± 1.0% vs. 48.7% ± 0.9% VO_2 peak, $P > 0.5$) in the two exercise trials were equivalent. In contrast, exercise before breakfast was performed with lower RQ, compared with that after breakfast (0.89 ± 0.01 vs. 0.94 ± 0.01, $P < 0.01$). When exercise was performed before breakfast, average rate of energy expenditure in the morning (0600–1200 h) was higher ($P < 0.01$), while that in the afternoon (1200–1800 h) was lower ($P < 0.01$) than those with exercise after breakfast trial. In the evening and during sleep, there was no significant difference in energy expenditure between the two trials. Average rate of fat oxidation was higher ($P < 0.01$) and carbohydrate oxidation was lower ($P < 0.01$) in the morning, when exercise was performed before breakfast. During the rest of the day, there were no significant differences in average rate of carbohydrate and fat oxidation between the two exercise conditions (Fig. 1). There was no significant difference in accumulated 24 h energy expenditure between the two trials (2594 ± 69 vs. 2587 ± 69 kcal/day, $P > 0.5$). Twenty four hour energy balance was slightly but significantly ($P < 0.05$) negative (− 136 ± 52 and − 129 ± 55 kcal/day), but the two exercise trials did not differ from each other ($P > 0.5$). Accumulated 24 h fat oxidation was more (720 ± 88 vs. 608 ± 82 kcal, $P < 0.05$) and carbohydrate oxidation was less (1543 ± 82 vs. 1669 ± 77 kcal/day, $P < 0.05$) when exercise was performed before breakfast. Urinary nitrogen excretion was not significantly different between the two trials (12.9 ± 1.3 vs. 12.1 ± 1.0 g/day, $P > 0.5$).

Apparent balance of energy, carbohydrate and fat in the morning was shifted downward by the exercise performed before breakfast compared with that of the exercise after breakfast trial. From lunch until the next morning, apparent balance of carbohydrate was shifted upward and that of fat was shifted downward in exercise before breakfast protocol, while energy balance in the two trials was similar (Fig. 2).

Activity counts increased when exercise with a bicycle ergometer was performed. Small increases in activity counts were also observed while subjects were eating meals, getting ready to sleep and out of bed. There was no significant difference in overall activity counts between the two trials (94 ± 7 vs. 98 ± 9 counts/min, $P > 0.5$) (Fig. 3).

4. Discussion

Exercise performed in post-absorptive state relied on fat oxidation (33.7% of total energy expenditure) more than that performed in postprandial state (19.1%), consistent with previous reports which compared energy metabolism during exercise performed in fasted and fed state [1–7]. The present study examined whether 24 h fat oxidation was affected by the time of exercise relative to breakfast, i.e. in post-absorptive and postprandial state. When exercise was performed in post-absorptive state, accumulated 24 h fat oxidation was more than that of exercise performed in postprandial

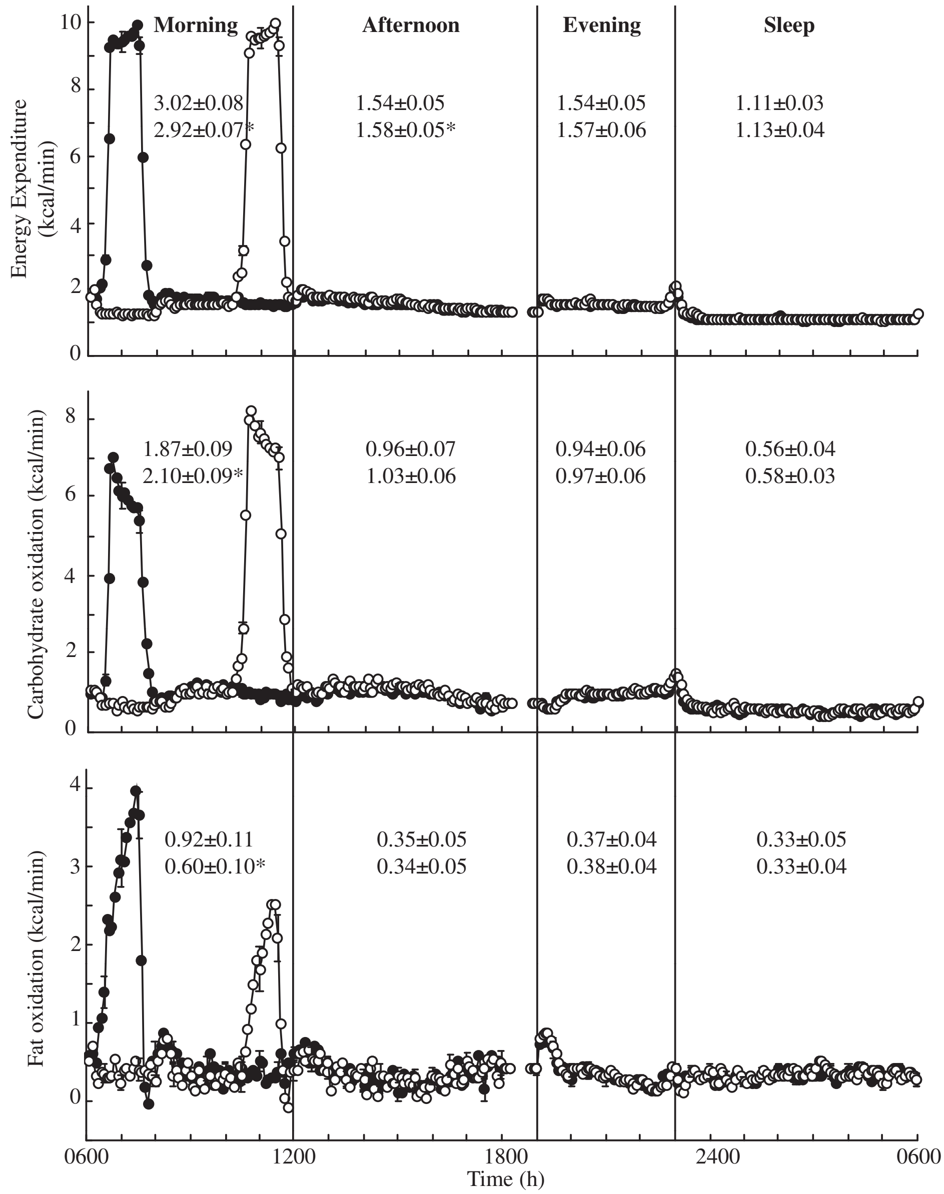

Fig. 1 – Effect of exercise before and after breakfast on 24 h energy expenditure and nutrients oxidation. Plots are means at every 5 min for exercise performed before (•) and after (○) breakfast conditions, and standard errors were shown only at every 30 min for clarity. Values of mean ± SE in morning (0600 h–1200 h), afternoon (1200 h–1900 h), evening (1900 h–2300 h) and sleep (2300 h–0600 h) were also shown for exercise performed before and after breakfast conditions. Significant differences between the two exercise conditions were shown as * ($P < 0.01$).

state. Our experimental design was strengthened by a prolonged indirect calorimetry until energy metabolism of the two experimental conditions became indistinguishable. During post-exercise recovery period, subjects were instructed to remain sedentary to reduce non-exercise activity thermogenesis, and physical activity during the 24 h calorimetry was confirmed to be similar in two trials.

Our findings on accumulated 24 h fat oxidation were at odds with what previous studies examining the effect of exercise at different intensity on 24 h substrate utilization suggested. The importance of post-exercise energy metabolism for 24 h nutrient balance was demonstrated in previous studies by Melanson et al., in which differences in fat oxidation during the exercise was offset by changes in metabolism during the post-exercise period, resulting in little difference in 24 h fat oxidation between experimental groups [13,21–23]. Exercise performed in post-absorptive state seems to have prolonged effects on substrate utilization through unique mechanisms. First, it is well established that decrease in endogenous and exogenous carbohydrate availability stimulates fat oxidation [24]. Exercise performed in post-absorptive state shifted the apparent carbohydrate balance downward (~ 350 kcal) for the first several hours of the day, compared with exercise in postprandial state. It is plausible that this short-term decrease in carbohydrate balance after the post-absorptive exercise continued to stimulate fat

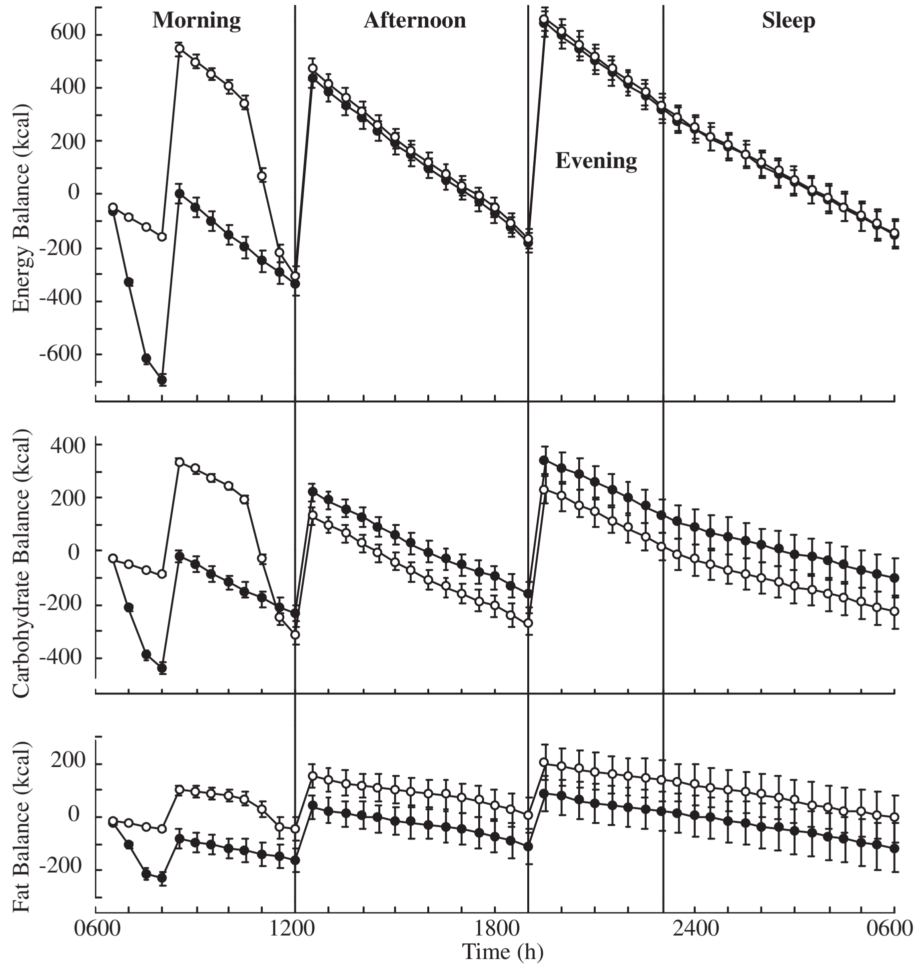

Fig. 2 – Effect of exercise before and after breakfast on energy and nutrient balance. Diurnal changes in energy, carbohydrate and fat balance were estimated as the difference between input and output. Setting initial reference value as 0 at 0600 h, mean values ± SE were plotted at every 30 min for exercise performed before (•) and after breakfast (○) conditions.

oxidation and prevented to offset the difference in fat oxidation between the two exercise conditions. Second, independently of changes in carbohydrate availability, an exercise-induced energy deficit per se also stimulates fat oxidation and suppresses carbohydrate oxidation. In the morning of the day after a single bout of exhaustive exercise, post-exercise increase in fat oxidation was greater in energy deficit (−1500 kcal/day) than in energy balanced condition [25]. Furthermore, the exercise-induced energy deficit up-regulated pyruvate dehydrogenase kinase, which inhibits carbohydrate oxidation by phosphorylating pyruvate dehydrogenase in muscle. Third, exercise after an overnight fast increased oxidation of dietary monounsaturated fat consumed shortly after the exercise [26,27]. Increased fat oxidation during the post-exercise period was accompanied by higher $^{13}CO_2$ recovery from dietary [1-^{13}C]oleate consumed after the exercise, although oxidation of dietary [d31]palmitate was not affected. Consistent with these metabolic studies, it has been reported that a single bout of exercise in the fasted state transiently up-regulated enzymes involved in fat oxidation such as LPL and carnitine palmitoyltransferase [28,29]. Thus, exercise performed in the post-absorptive state seems to prepare skeletal muscle suitable for fat oxidation. In the present study, experimental meals were prepared according to the dietary reference intakes for Japanese [15], fat content of which was comparatively lower than that of meal in other westernized countries [30]. A high fat meal, particularly that rich in monounsaturated fat, might enhance the stimulating effect of post-absorptive exercise on 24 h fat oxidation, and it remains to be examined.

In the present study, 24 h energy balance of both experimental conditions was slightly but significantly negative. It is worth mentioning that athletes are frequently in a negative energy balance, since training programs of athletes are periodized and highly varied over a week or block of weeks [31]. But it is unlikely that the present findings are only relevant to individuals aiming to reduce body fat by combining exercise and restricting energy intake. In the present study, the differences in 24 h fat oxidation between the two exercise trials were not correlated to those in energy balance ($r^2 = 0.002$, $P > 0.5$). As discussed already, exercise-induced negative energy balance increases fat oxidation [25]. However, exercise, independent of negative energy balance has little effect on accumulated 24 h fat oxidation, i.e. fat oxidation on

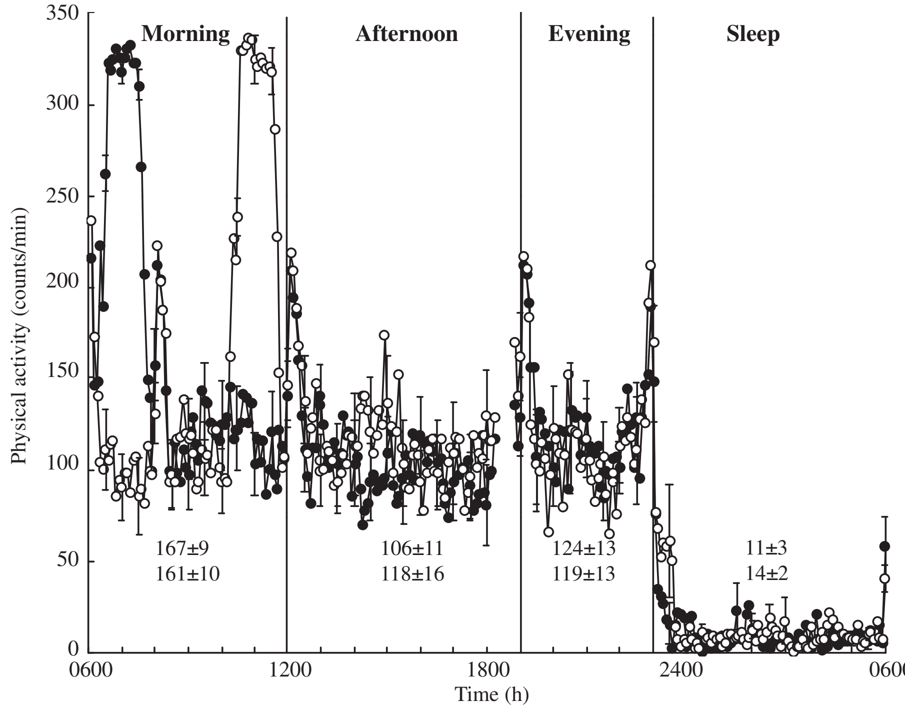

Fig. 3 – Effect of exercise before and after breakfast on gross motor activity. Plots are means at every 5 min for exercise performed before (●) and after (○) breakfast conditions, and standard errors were shown only at every 30 min for clarity. Values of mean ± SE in morning (0600 h–1200 h), afternoon (1200 h–1900 h), evening (1900 h–2300 h) and sleep (2300 h–0600 h) are also shown for exercise performed before and after breakfast conditions.

days with exercise is not different from that on sedentary control day in energy balanced condition [13,22,23]. Contrary to this current understanding, our findings suggest that exercise performed in the post-absorptive state increases accumulated 24 h fat oxidation. Unfortunately, resting control trial was not included in the present study, and it remains to be studied whether exercise performed in the post-absorptive state increases 24 h fat oxidation above sedentary control condition.

To evaluate translational potential of the present study, some considerations are required. First, carbohydrate intake before and during prolonged endurance exercise can improve endurance performance [32,33]. In contrast to this standard 'ergogenic' procedure, limiting carbohydrate availability during exercise further enhances training-induced adaptations in muscle to facilitate oxidative metabolism as well as fatty acid transport [3,34–38]. These recent findings are attracting athletes' attention to the "train-low, compete-high" approach, meaning train with low muscle glycogen levels to maximize the physiological adaptation and compete with high levels of glycogen stores [39]. The training in a glycogen-depleted state might stimulate protein catabolism, which may contradict the purpose of the training. However, in the present experimental condition, 24 h urinary excretion of nitrogen was not increased by exercise performed in the post-absorptive state. Secondly, although exercise performed in the post-absorptive state oxidized more fat, energy expenditure remained unchanged during the 24 h. It should be pointed out that energy expenditure over the entire day is more important than the source, i.e. carbohydrate or fat, when the effect of exercise on long-term body weight control is concerned. Due to the limited size of carbohydrate storage in the body, the findings of the present study can't be extrapolated to the chronic effects of the post-absorptive exercise. It is conceivable that the effect of post-absorptive exercise to expand glycogen storage would eventually be counterbalanced by increased carbohydrate oxidation. Alternatively, in free-living conditions, the expanded glycogen storage would inhibit subsequent energy intake, since carbohydrate balance is a strong predictor of subsequent ad libitum food intake [40,41]. Van Proeyen et al. demonstrated that early morning exercise in the fasted state was more potent than an identical amount of exercise in the fed state to prevent weight gain during hyper-caloric fat-rich diet [42]. In their study, energy intake of both exercise conditions was matched, but a possibility of under-reporting in food diary could not be denied.

In conclusion, an acute exercise bout in the post-absorptive state in the morning increased 24 h fat oxidation, relative to that performed in postprandial state. Further studies are needed to identify the sequence of events leading to body fat reduction, if exercise performed in post-absorptive state is beneficial to reduce body fat.

Author contributions

Conception and design of research were discussed among KS, YN and KT. Experiments were carried out by KS, YY, KI and KN. SY and MH analyzed data. Manuscript was drafted by KS and KT.

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Conflict of interest

All authors do not have conflicts of interest.

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