**Abstract** Indirect calorimetry with a room-size respiratory chamber provides an ideal setting to monitor energy metabolism for a prolonged period. During the last 10 years, experiments with this method have raised interesting observations such as: 1) exercise intensity has no effect on 24 h fat oxidation, 2) exercise has little, if any, effect on 24 h fat oxidation, and 3) exercise before breakfast increases 24 h fat oxidation. To some of the scientific community and general public, the first two statements may be unacceptable. But it can be factually explained that the impact of exercise on energy metabolism is not confined to the period of physical activity itself, and that fat oxidation remains elevated during the post-exercise period. The third observation seems to be insignificant, but contradicts the second one. It is premature to conclude that exercise has no effect on 24 h fat oxidation.

**Keywords**: human calorimeter, excess post-exercise oxygen consumption (EPOC), indirect calorimetry

**Introduction**

An indirect, yet accurate estimate of energy expenditure can be obtained by measuring oxygen uptake and carbon dioxide production. The principles of indirect calorimetry were described by Lavoisier in the 18th century, and from these early years, expired air has been collected using a mask (or mouth-piece), canopy (or hood), or chamber*. Without being hooked up with any apparatus, a metabolic chamber allows subjects to move freely within a small closed area. Whole room indirect calorimeter is a method for prolonged calorimetry, including during sleep, and for subjects who have difficulty being connected to a mask or hood, such as infants or small rodents (Fig. 1). During the last 10 years, continuous 24 h calorimetry with reasonable time resolution has raised interesting issues. This paper reviews the following three observations: 1) exercise intensity has no effect on 24 h fat oxidation, 2) exercise has little, if any, effect on 24 h fat oxidation, and 3) exercise before breakfast increases 24 h fat oxidation. To some of the scientific community and general public, the first two statements may be unacceptable; and the third observation seems to be insignificant, but contradicts the second one.

This paper is organized as follows. In section 1, after a brief explanation of the historical aspects, the theoretical basis of indirect calorimetry is explained. In section 2, the methodological details of the metabolic chamber are discussed. This section can be skipped without loss of context.

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Background and basics of the metabolic chamber

Historical aspect of the metabolic chamber. A respiration (metabolic) chamber for human indirect calorimetry was built by Pettenkoffer and Atwater et al. before the turn of the 20th century. Some experimental results reported by pioneers in this field stand the test of time. In 1919, Benedict et al. published their metabolic standard (BMR) from sex, age, height and weight\(^1\). As described in section 3, they reported, in 1910, an 11% increase in sleeping metabolic rate (SMR) as a result of exercise\(^2\). A review published in 1993 reported worldwide locations of modern metabolic chambers and 16 facilities were listed\(^3\). Chambers of the 20th century are equipped with an automated gas analyzer.

In Japan, a small coffin-like chamber to measure BMR was operating in the National Institute of Nutrition as early as 1921, according to personal communication from Dr. Shigeho Tanaka, National Institute of Health and Nutrition. Construction of a whole room calorimeter began in the year 2000 at the National Institute of Health and Nutrition; and it was followed by a limited, but rapidly increasing number of institutes particularly oriented to exercise physiology\(^4\) (Table 1).

Theoretical basis of indirect calorimetry. Theoretical details of indirect calorimetry have been described by Ferrannini\(^5\). Energy expenditure and macronutrient oxidation were calculated from oxygen uptake (\(\dot{V}\text{O}_2\)), carbon dioxide production (\(\dot{V}\text{CO}_2\)), and urinary nitrogen excretion. The rate of urinary nitrogen excretion (\(\dot{N}\)), an index of protein oxidation, was assumed to be constant during the urine collection period. Oxidation of glucose and fat were calculated as follows:

\(\dot{\text{Glucose oxidation (g/min)}} = 4.55 \dot{\text{VCO}}_2 \text{ (L/min)} - 3.21 \dot{\text{VO}}_2 \text{ (L/min)} - 2.87 \dot{N} \text{ (mg/min)} \) (1)

\(\dot{\text{Fat oxidation (g/min)}} = 1.67 \dot{\text{VCO}}_2 \text{ (L/min)} - 1.67 \dot{\text{VCO}}_2 \text{ (L/min)} - 1.92 \dot{N} \text{ (mg/min)} \) (2)

Once the rates of glucose, fat and protein oxidation have been computed, the total rate of energy production can be estimated by taking into account the caloric equivalent of the three substrates. The conversion factors for the calorific equivalents were 4.10 kcal/g protein (25.625 kcal/g urinary nitrogen), 9.50 kcal/g fat and 3.74 kcal/g carbohydrate. The equation for complete oxidation of glycogen is different from that of glucose, and is shown as follows:

\(\dot{\text{Glycogen (g/min)}} = 4.09 \dot{\text{VCO}}_2 \text{ (L/min)} - 2.88 \dot{\text{VCO}}_2 \text{ (L/min)} - 2.59 \dot{N} \text{ (mg/min)} \) (1)

Because there is relatively more carbon in 1 g of glycogen than in 1 g of glucose, the standard free-energy change for the oxidation of glycogen (4.1 kcal/g) is bigger than that of glucose (3.74 kcal/g). When there is reason to believe that tissue glycogen, rather than glucose, is the predominant form of carbohydrate being oxidized, it is appropriate to use equation (1)\(^1\). Having measured \(\dot{\text{VO}}_2\), \(\dot{\text{VCO}}_2\), and \(\dot{N}\), estimates of oxidized carbohydrate, using equation (1) and (1)\(^1\), will differ from each other. For example, from a particular set of \(\dot{\text{VO}}_2\) (0.40 L/min), \(\dot{\text{VCO}}_2\) (0.34 L/min) and \(\dot{N}\) (0.016 g/min) data, equations (1) and (1)\(^1\) give oxidation values of 0.217 g glucose/min and 0.197 g glycogen/min (-9.2%), respectively. However, the difference is negligible in terms of energy expenditure (0.812 kcal/min from (1) and 0.808 kcal/min from (1)\(^1\), a difference of only -0.4%). Thus, estimates of oxidized substrate are robust when expressed in kcal (not in g), even if type of carbohydrate oxidized in the body is unknown. Finally, it is worth mentioning that gross energy of glucose or glycogen for calculating energy expenditure should not be confused with the metabolizable energy of starch (4.0 kcal/g), which is referred to for calculation of energy intake.

Specification of whole room indirect calorimetry

Size and facilities. Currently in Japan, 9 institutes are operating 13 chambers, and sizes range from 14.5 to

<table>
<thead>
<tr>
<th>Year</th>
<th>Facility Name</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>2000</td>
<td>National Institute of Health and Nutrition</td>
<td>Japan</td>
</tr>
<tr>
<td>2002</td>
<td>Doctoral Program in Sports Medicine, University of Tsukuba</td>
<td>Japan</td>
</tr>
<tr>
<td>2003</td>
<td>Health Care Food Research Laboratories, Kao Corporation</td>
<td>Japan</td>
</tr>
<tr>
<td>2005</td>
<td>Department of Sport and Exercise Nutrition, Sendai University</td>
<td>Japan</td>
</tr>
<tr>
<td>2009</td>
<td>Graduate School of Health and Sports Science, Doshisha University</td>
<td>Japan</td>
</tr>
<tr>
<td>2009</td>
<td>Department of Social Medicine, Hirosaki University School of Medicine</td>
<td>Japan</td>
</tr>
<tr>
<td>2010</td>
<td>Faculty of Health and Sports Science, Fukuoka University</td>
<td>Japan</td>
</tr>
<tr>
<td>2010</td>
<td>Graduate School of Sport and Health Science, Ritsumeikan University</td>
<td>Japan</td>
</tr>
<tr>
<td>2011</td>
<td>School of Health and Sports Science, Juntendo University</td>
<td>Japan</td>
</tr>
</tbody>
</table>
20 m³. The largest chamber in the world is 34 m³, and is located at the USDA Children’s Nutrition Research Center in Houston, USA⁶. Standard facilities include a toilet, bed, desk, chair and phone. Usually, a shower is not installed in the chamber to avoid technical challenges caused by rapid changes in moisture, which potentially causes error in gas analysis and control of flow rate of incoming and outgoing air. The exception is a chamber in Göteborg, Sweden, which is equipped with a shower⁷. A bicycle ergometer or treadmill can be put in the chamber for exercise physiology. Any apparatus that doesn’t consume O₂ or produce CO₂ can be used in the chamber. Polysomnography (PSG) is used to monitor, and light-emitting diodes (LEDs) of short wavelength are used to interfere with sleep.

**Analyzer.** Efforts were made to improve our experimental setting above the "global standards". Standard whole room metabolic chambers around the world are equipped with differential pressure paramagnetic analyzers for O₂ and infrared absorption photometers for CO₂ concentration measurement, respectively. The standard deviation (SD) of continuous measurement for standard gases is 0.010% for O₂ and 0.004% for CO₂, for the paramagnetic and infrared analyzer, respectively, according to manufacturer specifications⁸. The former is less accurate and limits overall accuracy of calorimetry. Mass spectrometers, particularly the magnetic sector type rather than quadrupole type, adopted by the majority of Japanese metabolic chambers, are extremely accurate and stable. For example, mass spectrometer SDs, for the continuous measurement of calibration gas mixtures, are 0.001 and 0.0003% for O₂ and CO₂, respectively, for a recent sector type mass spectrometer model (Prima Pro, Thermo Fisher Scientific, Winsford, UK). Traditionally, O₂ and CO₂ concentrations in the air were measured, and the rest of the components were assumed to be inert gasses such as N₂ and argon. By measuring all major components of an air sample (O₂, CO₂, 13CO₂, N₂ and argon), we can suppress the drift effect of the analysis during prolonged measurements.

**Algorithm.** Whole room indirect calorimetry provides accurate measurement of energy metabolism over long periods of time; but it has limitations to assess dynamic changes. Improvements in noise reduction algorithms have made the whole body calorimeter useful, not only for traditional 24-h energy expenditure measurements, but also for experiments requiring rapid response. Several noise reduction algorithms have been proposed⁷⁻¹¹; and rigorous comparison among the algorithms, under various conditions, rated the deconvolution with regularization parameter as the best, and Kalman smoothing as a close second. Both the trends identification and moving average algorithms have limited ability to capture transient response¹¹ (Fig. 2). With the right combination of a sensitive gas analyzer and good algorithm, changes in sleeping metabolic rate and RQ to sleep stages could be accurately determined¹².

**EPOC**

In 1910, Benedict and Carpenter reported an 11.1% increase in energy expenditure during sleep, which was 7-13 hours after severe exercise². The prolonged effects of exercise on energy metabolism have been confirmed by subsequent studies¹³,¹⁴. The classical “oxygen debt” hypothesis in the 1920s explained that elevated oxygen consumption after exercise was necessary for the repayment of the oxygen deficit incurred after the start of exercise and the oxidative removal of lactate. However, subsequent studies revealed that the time frame of lactate removal and post-exercise oxygen consumption were not causally related; and the term ‘excess post-exercise oxygen consumption’ (EPOC) was coined to avoid the implication of causality⁴. The magnitude of EPOC after aerobic exercise depends on its intensity, and an intensity above 50-60% of VO₂ max is required in order to induce EPOC lasting for several hours after exercise⁶. At exercise intensities at or above this level, a linear relationship between the magnitude of EPOC and the duration of the exercise bout was observed. One of the most exhaustive bouts of exercise in any of the EPOC studies was a full marathon, with an intensity and duration of 65 to 80% of VO₂ max for 2 to 3 h, respectively¹⁷,¹⁸. Touminen et al reported a 6% increase in basal metabolic rate (BMR) on the morning after the marathon³⁹. An estimate, made by the authors, of prolonged EPOC after a marathon race was 152 kcal at 22 h³⁹. Although some researchers claimed the possibility that more energy would be expended following a bout of exercise than during the exercise itself⁴⁰, the estimated magnitude of EPOC after a marathon was small when compared to the estimated 2500 kcal energy expenditure during the actual race³⁹. When low-intensity physical activities were intermittently repeated, EPOC was negligible⁴¹.

In addition to enhanced oxygen consumption, a relative shift from carbohydrate to fat as substrate source is a consistent finding after prolonged exhaustive exercise¹³,¹⁸,²²-²⁵. A single exercise bout can rapidly increase fat oxidation for up to 36 h following the exercise period³⁵,²⁶,²⁷. As observed after a marathon race or laboratory experiment, the time course of EPOC and the respiratory quotient (RQ) can be dissociated, suggesting that different mechanisms are responsible for the two processes¹³,¹⁸ (Fig. 3). Cumulative fat oxidation during 14 h (7-21 h after the race including a sleeping period) doubled after a marathon race (27.2 ± 1.4 g/14 h vs 50.5 ± 2.8 g/14 h, P<0.01)³⁹. Although the amount of fat oxidized during a given post-exercise period is not spectacular, it becomes decisive when the effect of exercise intensity on 24 h fat balance is evaluated.
Effect of exercise intensity on 24 h fat oxidation

Substrate oxidized during exercise depends on exercise intensity. For low-intensity exercise, below 40–50% $V_{\text{O}2\text{max}}$, the energy supplied is primarily from oxidation of plasma free fatty acids. As exercise intensity increases, additional energy is obtained by the utilization of muscle glycogen, blood glucose and intramuscular triglyceride. During high-intensity exercise, fat oxidation gradually decreases until 100% carbohydrate oxidation occurs at about 100% of $V_{\text{O}2\text{max}}$. This has led to the commonly held view that low-intensity exercise may be better than high-intensity exercise for increasing fat oxidation and, thus, weight loss. However, the impact of exercise on energy metabolism is not confined to the period of physical activity itself, and post-exercise changes in energy metabolism should also be considered in evaluating the effect of exercise on 24-h nutrient balance. Comparison of fat oxidation during the post-exercise recovery period following isocaloric exercise performed at high (65% $V_{\text{O}2\text{max}}$ for 1 h) or moderate intensity (45% $V_{\text{O}2\text{max}}$ for 86-89 min) revealed the following. First, consistent with the literature, fat oxidation during exercise was less for high-intensity exercise than for low-intensity exercise.
Second, the increase in fat oxidation during the post-exercise period seemed to be greater after high-intensity exercise, although the difference did not reach statistical significance. Third, the sum of fat oxidation during the exercise and post-exercise periods was not significantly different between the two exercise conditions of different intensity. Fourth, the increase in fat oxidation during the post-exercise period is greater in male than in female subjects, consistent with sex differences in lipolysis and fatty acid metabolism during the post-exercise period. Although females are capable of deriving a great portion of energy expenditure from fat oxidation during exercise, they are typically less successful in achieving the goal of fat loss in response to exercise interventions. This apparent paradox can be explained by substantial sex differences in fat metabolism during the post-exercise recovery period. The relatively short observation period, collecting expired air by mouthpiece, leaves uncertainties as to whether any difference in fat oxidation during exercise is offset by changes in fat oxidation during the remainder of the day.

The concept that “low-intensity exercise is best for maximizing fat oxidation and fat loss” has been challenged, during the last 10 years, by studies on 24-h energy metabolism using a metabolic chamber. A series of experiments has been carried out under an energy balanced condition (i.e., energy intake and expenditure are nearly equal). Melanson et al. compared 24-h fat oxidation after isocaloric exercise (400 kcal) performed at 40% or 70% VO₂ max. During exercise, fat oxidation at low intensity was greater than that of high-intensity exercise (85.2 ± 4.0 g vs 21.4 ± 1.8 g, P<0.01), but post-exercise 24 h fat oxidation was unaffected by exercise intensity (Fig. 5). The results were confirmed in their other experiments with other sets of subjects, and consistent with reports by other research groups using a metabolic chamber (Table 2). Melanson et al. implied that “Given that time is a limiting factor for most individuals, if the goal of exercise is to maximize fat oxidation to better regulate body fat mass, then exercise should be performed at the highest intensity that can be comfortably maintained.” These data suggest that suppressed fat oxidation during higher intensity exercise is offset during the post-exercise period. However, due to the time resolution of whole room indirect calorimetry used in the previous studies, there is a paucity of literature showing the time course of fat oxidation to reveal when fat oxidation catches up after exercise.

The series of studies also implies the intuitively unacceptable message that “exercise doesn’t enhance 24 h fat oxidation when the energy balance is maintained” as discussed below.

Effect of exercise on 24 h fat oxidation

Since fat oxidation is increased by exercise, it is assumed by many, in the scientific community and the general public, that more fat is oxidized on days when exercise is performed. However, a series of studies on the independent effects of exercise on 24-h fat oxidation demonstrated that fat oxidation on days with exercise doesn’t differ from sedentary control days when the energy balance is maintained.

Unlike “standard” exercise physiology in which subjects are kept in a fasted state during exercise and the recovery period, it is natural to provide meals to subjects...
during prolonged calorimetry in a metabolic chamber. A positive (negative) energy balance suppresses (enhances) fat oxidation. For example, an exercise-induced negative energy balance acts as a driving force to increase fat oxidation. On the morning of the day after a single bout of exhaustive exercise, fat oxidation was enhanced and carbohydrate oxidation was suppressed. Interestingly, these effects of exercise on energy metabolism of the next morning, were greater in an energy deficit condition than in an energy balanced condition. Among macronutrients, carbohydrate overfeeding yields an immediate and profound shift in fuel use, suppressing the oxidation of fat and increasing carbohydrate oxidation and energy expenditure. In contrast, fat is not effective in promoting oxidation during overfeeding and also does not stimulate an increase in energy expenditure. Therefore, a macronutrient-balanced condition is ideal for assessing the effect of exercise on 24-h fat oxidation independent of energy and carbohydrate balance.

The independent effects of exercise on 24-h fat oxidation have been assessed under an energy-balanced condition. As listed in table 2, experimental results are unanimous, and don’t support the notion that “exercise increases 24-h fat oxidation” under an energy-balanced condition. However, it is possible that these studies suffer from type II errors due to the limited sensitivity of whole room indirect calorimetry and the small number of subjects. As shown in fig. 5, ANOVA didn’t detect the main effects of an exercise bout on 24-h fat oxidation, but mean values of exercising days tended to be higher than those of sedentary days in male and female.

Table 2. Effect of exercise bout and its intensity on 24 h fat oxidation.

<table>
<thead>
<tr>
<th>Institute</th>
<th>Subject</th>
<th>Exercise Intensity</th>
<th>Duration</th>
<th>Effect of Exercise</th>
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<tr>
<td>Lausanne</td>
<td>50%</td>
<td>60min</td>
<td>increase</td>
<td></td>
</tr>
<tr>
<td>Laval</td>
<td>50%</td>
<td>60min</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>Maastricht</td>
<td>38%</td>
<td>60min</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>Colorado</td>
<td>45%</td>
<td>~100min</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td></td>
<td>70%</td>
<td>~60min</td>
<td>no</td>
<td></td>
</tr>
<tr>
<td>Colorado</td>
<td>obese</td>
<td>control</td>
<td>40%</td>
<td>60min</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>70%</td>
<td>40min</td>
</tr>
<tr>
<td></td>
<td>lean</td>
<td>control</td>
<td>40%</td>
<td>60min</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>70%</td>
<td>40min</td>
</tr>
<tr>
<td>Colorado</td>
<td>young</td>
<td>control</td>
<td>60%</td>
<td>34min</td>
</tr>
<tr>
<td></td>
<td>old</td>
<td>control</td>
<td>60%</td>
<td>62min</td>
</tr>
<tr>
<td>Colorado</td>
<td>control</td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>lean sedentary</td>
<td>55%</td>
<td>60min</td>
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</tr>
<tr>
<td></td>
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<td>55%</td>
<td>60min</td>
<td>no</td>
</tr>
<tr>
<td></td>
<td>old sedentary</td>
<td>55%</td>
<td>60min</td>
<td>no</td>
</tr>
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</table>

Independent effects of exercise intensity (low vs high intensity) and exercise itself (control vs exercise) on 24 h fat oxidation have been assessed under energy-balanced study design, except one study at Lausanne, in which subjects were in a state of negative energy balance. no: not significantly different.

Effect of exercise before or after breakfast on nutrient oxidation

Fat oxidation during exercise is suppressed by carbohydrate ingestion before exercise. This effect persists for at least 4 h after a meal, based on studies comparing metabolic response to exercise performed 2, 4, 6, 8 and 12 h after carbohydrate ingestion. For individuals taking 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,
common practice among athletes and recreational runners, is beneficial to reduce body fat. However, as discussed in a previous section, it is necessary to take the post-exercise period into account, when the effect of exercise on body fat balance is concerned. A few studies extended their observations on energy metabolism into the post-exercise period. Compared with exercise after breakfast, exercise performed before breakfast oxidized more\(^{48}\) or less fat\(^{41,49}\) during a 2 to 6 h post-exercise period. Thus, experimental results were inconsistent, and the experimental protocol in these studies didn’t allow interpretation of the results to be straightforward. For example, the duration of the post-prandial and post-exercise period was not matched in the two exercise protocols, when the timing of exercise and breakfast was swapped. Also, indirect calorimetry was terminated before energy metabolism of the two exercise conditions became indistinguishable, suggesting that EPOC and/or the thermic effect of food were still ongoing at the end of the experiment. Thus, it is still inconclusive whether exercise before breakfast increases 24-h fat oxidation.

The authors compared the effects of exercise at 50\% VO\(_2\) max for 60 min, performed before and after breakfast, on 24 h energy expenditure and nutrient oxidation\(^{50}\). The 24-h energy expenditure was not affected by time of exercise relative to breakfast (exercise before breakfast 2594 ± 69 vs exercise after breakfast 2587± 69 kcal, P>0.5). Furthermore, 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, P<0.05) when exercise was performed before breakfast (Fig. 6).

The time course of “apparent” energy and nutrient

![Graph showing energy expenditure and nutrient oxidation over 24 hours](image)

Fig. 6  Effect of exercise before breakfast or after breakfast on 24 h energy expenditure and nutrients oxidation.

Plots are means at every 5 min for exercise before (●) and after (○) breakfast conditions, and standard errors were shown at every 30 min. Values of mean ± SE during morning (0600h-1200h), afternoon (1200h-1900h), evening (1900h-2300h) and sleep (2300h-0600h) are also shown for exercise before and after breakfast conditions. Repeated measures two-way ANOVA revealed significant main effect of time on energy expenditure, carbohydrate oxidation and fat oxidation (P<0.01). Main effect of exercise condition was significant on carbohydrate and fat oxidation (P<0.05) but not on energy expenditure (P>0.5). Interaction of time and exercise condition was significant on energy expenditure, carbohydrate oxidation, and fat oxidation (P<0.01). Significant differences between the two exercise conditions by multiple comparisons were shown as * (P<0.01)\(^{50}\).
balance revealed that the energy balance shifted downward by about 500 kcal until lunch, when exercise was performed before breakfast (Fig. 7). It is likely that an exercise-induced energy deficit acted as the driving force to increase fat oxidation. Secondly, exercise performed before breakfast burned more glycogen and stimulated non-oxidative carbohydrate storage of breakfast, which resulted in suppressed carbohydrate oxidation and enhanced fat oxidation.

The results of this study demonstrate that exercise before breakfast could be regarded as a useful tool to reduce body fat; but the chronic effects of exercise before breakfast remain to be studied. In preparing for prolonged fasting during sleep, the body’s energy balance shifts in a positive direction during the day; and the energy status of people with a normal lifestyle reaches its zenith after supper. The population that exercises after supper seems to be increasing due to changes in lifestyle. But data on the continuous measurement of energy metabolism in a metabolic chamber suggest that exercise after supper is a much less effective practice to reduce body fat.

Scientific and practical implications

EPOC is a complex entity, and its magnitude after ordinary exercise was not as spectacular as initially claimed. However, when the daily energy balance of one’s body is near equilibrium, EPOC can be decisive in tilting the delicate balance of energy and nutrients in a positive or negative direction. If the effect of exercise on 24-h fat oxidation depends on when it is performed, as discussed in section 6, one has to be cautious in accepting the notion that “exercise has little effect on 24-h fat oxidation”. Unfortunately, a sedentary control was not included in the above study, so it remains to be evaluated whether or not exercise performed before breakfast increases 24-h fat oxidation.

Although this review didn’t give more consideration to sex differences in 24-h fat metabolism, the literature supports the differences between male and female subjects in fat metabolism during the post-exercise period. And, although it is time consuming, 24-h indirect calorimetry is an essential tool for studying the long-term effects of exercise on substrate oxidation and energy expenditure.

![Fig. 7](image-url)  **Effect of exercise before breakfast or after breakfast on energy and nutrient balance.** Diurnal rhythm of energy, carbohydrate and fat balance were estimated as difference between input and output. Setting initial reference value as 0 at 0600h, mean values ± SE were plotted at every 30 min for exercise before breakfast (●) and exercise after breakfast (○) conditions.
However, since the number of metabolic chambers is limited among institutes, it is recommended that efficient multicenter studies are utilized.

Note
*Metabolic chamber, whole room indirect calorimeter and human calorimeter are interchangeably used in this review. Strictly speaking, a metabolic chamber refers to an air-tight space, and whole room indirect calorimeter or human calorimeter refers to a system that consists of a metabolic chamber and gas analyzer.

References
1) McArdle WD, FI Katch, VL Katch. 1999. Sports & Exercise Nutrition. pp 344-345, Lippincott Williams & Wilkins, Maryland, USA.
polysis and fatty acid metabolism in men and women during the postexercise recovery period. *J Physiol* 584: 963-981.