# Determination of the exercise intensity that elicits maximal fat oxidation

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#### ABSTRACT

ACHTEN, J., M. GLEESON, and A. E. JEUKENDRUP. Determination of the exercise intensity that elicits maximal fat oxidation. Med. Sci. Sports Exerc., Vol. 34, No. 1, 2002, pp. 92-97. Purpose: The aim of this study was to develop a test protocol to determine the exercise intensity at which fat oxidation rate is maximal (Fat<sub>max</sub>). Method: Eighteen moderately trained cyclists performed a graded exercise test to exhaustion, with 5-min stages and 35-W increments (GE<sub>35/5</sub>). In addition, four to six continuous prolonged exercise tests (CE) at constant work rates, corresponding to the work rates of the GE test, were performed on separate days. The duration of each test was chosen so that all trials would result in an equal energy expenditure. Seven other subjects performed three different GE tests to exhaustion. The test protocols differed in stage duration and in increment size. Fat oxidation was measured using indirect calorimetry. Results: No significant differences were found in  $Fat_{max}$  determined with the  $GE_{35/5}$ , the average fat oxidation of the CEtests, or fat oxidation measured during the first 5 min of the CE tests ( $56 \pm 3$ ,  $64 \pm 3$ ,  $58 \pm 3\% \text{VO}_{2\text{max}}$ , respectively). Results of the GE35/5 protocol were used to construct an exercise intensity versus fat oxidation curve for each individual. Fat<sub>max</sub> was equivalent to  $64 \pm 4\% \dot{V}O_{2max}$  and  $74 \pm 3\% HR_{max}$ . The Fat<sub>max</sub> zone (range of intensities with fat oxidation rates within 10% of the peak rate) was located between 55  $\pm$  3 and 72  $\pm$  4%VO<sub>2max</sub>. The contribution of fat oxidation to energy expenditure became negligible above 89  $\pm$ 3%VO<sub>2max</sub> (92 ± 1%HR<sub>max</sub>). When stage duration was reduced from 5 to 3 min or when increment size was reduced from 35 to 20 W, no significant differences were found in Fat<sub>max</sub>, Fat<sub>min</sub>, or the Fat<sub>max</sub> zone. Conclusion: It is concluded that a protocol with 3-min stages and 35-W increments in work rate can be used to determine Fat<sub>max</sub>. Fat oxidation rates are high over a large range of intensities; however, at exercise intensities above  $Fat_{max}$ , fat oxidation rates drop markedly. **Key Words:** EXERCISE TESTING, FAT METAB-OLISM, INDIRECT CALORIMETRY, CYCLING

▶ hifts in energy substrate mobilization and utilization occur as exercise intensity increases (22). There is a progressive increase in the relative contribution of carbohydrate oxidation to energy expenditure and a corresponding decrease in the relative contribution of fat oxidation to energy expenditure. However, from low to moderate intensities of exercise, the absolute rate of fat oxidation increases and then declines as exercise becomes even more intense (5,20,22,28). Several mechanisms have been proposed to explain the lower fat oxidation rates at high compared with moderate exercise intensities. Fatty acid oxidation during exercise may be controlled to some extent by the availability of plasma free fatty acids (6,28), and also at the muscle tissue level, where entry of fatty acyl-CoA into the mitochondria is the rate-limiting step in fat oxidation (6,30) (for review see (17–19)).

Treatments that prevent conditions like overweight and obesity are of considerable interest both to the general public and health-care professionals (31). Probably the most important of these treatments is regular exercise that increases daily energy expenditure and fat oxidation. In addition, it has been shown in athletes that after endurance training, fat oxidation at a given

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intensity is increased that coincides with increases in performance (13,14). These observations indicate that the ability to oxidize fatty acids is related to improved performance. These changes are likely to be the results of an overall increased aerobic capacity (19).

Generally, the highest rates of fat oxidation are found at low to moderate exercise intensities (range 33–65%  $\dot{V}O_{2max}$ ) (1,2,4,11,16,20,28–31). Most studies, however, measured fat oxidation at only two (1,4,11,20,30,31), three (28,29), or four (2,16) different exercise intensities. This makes it difficult to accurately determine the exercise intensity that elicits maximal fat oxidation. To our knowledge, there are no papers in the literature that have systematically studied fat oxidation over a large range of exercise intensities to identify the exercise intensity at which fat oxidation is maximal.

Therefore, the main purpose of this study was to develop an exercise protocol to determine the exercise intensity that elicits the maximum fat oxidation rate.

#### **METHODS**

**Subjects.** Twenty healthy, moderately trained men participated in this study, which was approved by the South Birmingham Local Research Ethics Committee of the University of Birmingham, UK. Each volunteer gave his written informed consent after explanations of the experimental procedures and possible risks and benefits. The subjects

TABLE 1. Physical and physiological characteristics of subjects.

				VO <sub>2max</sub>		W <sub>max</sub>	
N	Age (yr)	Body Mass (kg)	Body Fat (%)	(L·min <sup>-1</sup> )	(ml·min <sup>-1</sup> ·kg <sup>-1</sup> )	(W)	(W·kg <sup>-1</sup> )
18	28 = 2	74.8 ± 1.5	$16.2 \pm 1.2$	$4.3 \pm 0.1$	58.4 ± 1.8	348 ± 7	$4.7 \pm 0.1$
7	26 = 3	75.3 ± 0.9	11.5 ± 0.8	4.8 ± 0.1	64.2 ± 1.6	386 = 11	5.1 ± 0.2

Values: mean ± SEM.

were all club/country standard endurance cyclists with a training background of at least 3 yr. The characteristics of the subjects are shown in Table 1.

General design. Eighteen subjects performed a graded exercise (GE) test to exhaustion on a cycle ergometer; the results of this test were used to measure fat oxidation over a wide range of intensities for each subject. To test whether this graded exercise protocol could be used to accurately determine the exercise intensity that elicits maximal fat oxidation (Fat<sub>max</sub>), the subjects performed a set of four to six additional continuous prolonged exercise (CE) tests on separate days at exercise intensities corresponding to those during the GE test. The duration of the CE tests was decreased with increasing work rates to elicit similar energy expenditure over the trials. For each subject, fat oxidation rate was plotted against exercise intensity to construct a fat oxidation curve. The fat oxidation curve of each CE test was compared with the curve of the GE test. To investigate whether stage duration and work rate increment size affected the results, in a second study, seven subjects were asked to perform three additional GE tests with different exercise protocols. All abbreviations used in the text to refer to different test protocols are defined in Table 2.

Experimental design. Maximal oxygen uptake, maximal work rate, and maximal fat oxidation ( $\dot{V}O_{2max}$ ,  $W_{max}$ ) and Fat<sub>max</sub>) were determined in 18 subjects by using a GE test to exhaustion on an electromagnetically braked cycle ergometer (Excalibur, Lode, Groningen, The Netherlands). Before the start of the experiments, the subjects were familiarized with the equipment and the procedures. Experiments were always performed in the morning (start of exercise between 8 and 10 a.m.) and at the same time to avoid circadian variance. Subjects were asked to fill in a 1-d food diary on the day before their first test, and they were asked to repeat this diet before all subsequent trials. Furthermore, subjects were asked to avoid strenuous exercise the day before the test. Subjects reported to the laboratory after a 10to 12-h overnight fast, and body mass and height were determined. Body fat was estimated from skin-fold thickness measurements at four sites according to the methods of Durnin and Womersley (7). A Teflon catheter (Quickcath, Baxter, Norfolk, UK) was introduced into an antecubital arm vein and connected to a three-way stopcock (Sims Portex, Kent, UK), and an initial blood sample was collected. The catheter was maintained patent with isotonic

saline (Baxter, Norfolk, UK). Subjects started cycling at 95 W, and the work rate was increased by 35 W every 5 min. When the respiratory exchange ratio (RER) reached 1.0, the work rate was increased by 35 W every 2 min until exhaustion. During the last 30 s of each exercise intensity, a blood sample (~1 mL) was collected. Heart rate was recorded continuously during the test by using a radio telemetry heart rate monitor (Polar Vantage NV, Polar Electro Ltd., Ov, Finland). Breath-by-breath measurements were performed throughout exercise using an Oxycon Alpha (Jaeger, Würzberg. Germany) gas analysis system. The volume and gas analyzers of the system were calibrated using a 3-L calibration pump and calibration gas (15.12% O<sub>2</sub>; 5.10% CO<sub>2</sub>), respectively. Maximal work rate was calculated from the last completed work rate plus the fraction of time spent in the final noncompleted work rate multiplied by the work rate increment. A levelling off of oxygen uptake (defined as an increase of no more than 2 mL·kg<sup>-1</sup> body weight<sup>-1</sup>·min<sup>-1</sup>) during the latter stages of the exercise test was taken to indicate that maximal oxygen uptake had been attained.  $\dot{V}O_{2max}$  was calculated as the average oxygen uptake of the last 60 s of the test.

The subjects performed a set of additional tests (CE tests) at the exercise intensities corresponding to those during the GE test where RER < 1. The duration of each test was chosen so that all trials would result in an equal total energy expenditure of 2.8 MJ. The CE-test duration ranged from 80 to 35 min for workloads ranging from 95 to 270 W, respectively. The order of the tests was randomized and separated by at least 2 d. During the tests, respiratory gas exchange measurements were performed during the first 10 min of exercise and during 5-min intervals thereafter. Only data collected during the last 3 min were used.

In an attempt to find a more practical but still valid protocol, seven subjects were asked to perform three additional graded exercise tests to exhaustion (the characteristics of this subgroup are displayed in Table 1). The exercise protocol performed was different during each test. The three protocols were as follows:  $GE_{35/5}$ : 35-W increments every 5 min until the RER reached 1.0, after which the work rate was increased by 35 W every 2 min until exhaustion;  $GE_{35/3}$ : 35-W increments every 3 min until exhaustion; and  $GE_{20/3}$ : 20-W increments every 3 min until exhaustion. All tests started at 95 W. The order of the tests was randomized and the tests were at least 2 d apart. All tests were performed

TABLE 2. Graded and continuous exercise tests.

GE <sub>35.5</sub>	Graded exercise test to exhaustion: start 95 W. 5-min stages with 35-W increments till RER = 1.0 hereafter 2-min stages
GE <sub>35-3</sub>	Graded exercise test to exhaustion: start 95 W. 3-min stages with 35-W increments
GE <sub>20:3</sub>	Graded exercise test to exhaustion: start 95 W. 3-min stages with 20-W increments
CE <sub>average</sub>	Continuous exercise test at work rates corresponding to work rates of GE <sub>35/5</sub> test: Fat oxidation calculated over entire test
CE <sub>5-min</sub>	Continuous exercise test at work rates corresponding to work rates of GE <sub>35.5</sub> test: Fat oxidation calculated over first 5 minutes of test

in the morning after an overnight fast. During all three tests, respiratory gas exchange was measured and heart rate was recorded continuously.

Indirect calorimetry and calculations. During all GE tests, average values for  $\dot{V}O_2$  and  $\dot{V}CO_2$  were calculated over the last 2 min of every stage. During CE tests, the values during the last 3 min of each 10-min interval (i.e., 7–10 min; 17–20 min; etc.) were averaged. These values were then again averaged to obtain one set of values per CE test ( $CE_{average}$ ). In addition,  $\dot{V}O_2$  and  $\dot{V}CO_2$  were averaged from min 3 to 5 during every CE test ( $CE_{5-min}$ ) (similar to calculations in GE tests). Fat and carbohydrate oxidation and energy expenditure were calculated using stoichiometric equations (10) and appropriate energy equivalents, with the assumption that the urinary nitrogen excretion rate was negligible. Fat and carbohydrate oxidation were thus obtained for GE,  $CE_{average}$ , and  $CE_{5-min}$ .

For each individual, the results of the GE test were used to construct a curve of fat oxidation rate versus exercise intensity, expressed as  $\dot{V}O_2$  and HR. The curve was used to determine the following variables:

Fat<sub>max</sub>: the exercise intensity at which the highest rate of fat oxidation was observed.

Fat<sub>min</sub>: the exercise intensity where the fat oxidation rate reached zero (i.e., where RER  $\geq 1.0$ ).

 $Fat_{max}$  zone: range of exercise intensities with fat oxidation rates within 10% of fat oxidation rates at  $Fat_{max}$ . The lower limit will be referred to as "low" and the upper limit as "high."

 ${\rm Fat_{max}}$  was determined using data from GE,  ${\rm CE_{average}}$ , and  ${\rm CE_{5-min}}$ . If the cycling economy of the subjects during a stage of the GE test or during one of the CE tests was below an arbitrary 3.35 kJ·L  ${\rm O_2}^{-1}$  (average economy of GE tests minus 2 standard deviations), the fat oxidation rates at that stage were not taken into consideration for the  ${\rm Fat_{max}}$  (-zone) determination. Here cycling economy is defined as the amount of energy transferred to the cycle per L of oxygen consumed (24).

To quantify Fat<sub>max</sub> and to compare the three different exercise protocols, the results of the GE tests were used to compose an average fat oxidation curve. For each individual Fat<sub>max</sub>, Fat<sub>min</sub>, and exercise intensities that elicited fat oxidation rates 5, 10, and 20% below the peak rate were determined. These specific points were then averaged and plotted against the average fat oxidation rates.

**Statistical analysis.** Experimental data are presented as means  $\pm$  SEM. Significant differences in Fat<sub>max</sub>, Fat<sub>min</sub>, and the fat oxidation rate at Fat<sub>max</sub> between GE, CE<sub>average</sub>, CE<sub>5-min</sub>, and the maximal performance between the GE<sub>35/5</sub>, GE<sub>35/3</sub>, and GE<sub>20/3</sub> tests were identified by use of a one-way analysis of variance. A general linear model for repeated measures was used to identify differences between the fat oxidation curves of GE<sub>35/5</sub>, GE<sub>35/3</sub>, and GE<sub>20/3</sub>. When a significant *F*-ratio was obtained, the Tukey *post hoc* test was used to compare means. For all statistical analyses, significance was accepted at P < 0.05.

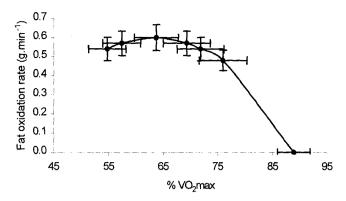


FIGURE 1—Fat oxidation rates versus exercise intensity expressed as percentage of  $\dot{VO}_{2max}$ ; N = 11; values are mean  $\pm$  SEM.

### **RESULTS**

Figure 1 shows the relationship between fat oxidation rate and exercise intensity, expressed as a percentage of maximal oxygen uptake. To construct this curve, the results of the GE test of 11 subjects of the first study were used. For the remaining seven subjects, there were insufficient data points to construct the curve (i.e., there were no or insufficient intensities below Fat<sub>max</sub>). The exercise intensity below Fat<sub>max</sub> at which fat oxidation rates were 20% below fat oxidation at Fat<sub>max</sub> could only be determined in five subjects and was therefore not included in Figure 1.

With increasing exercise intensities, the fat oxidation rate increased to a maximum of  $0.60\pm0.07~{\rm g\cdot min}^{-1}$  at  $64\pm4\%{\rm VO}_{2\rm max}$  (range 42-84), which corresponds to  $74\pm3\%{\rm HR}_{\rm max}$  (range 54-92). The intensities of the  ${\rm Fat}_{\rm max}$  zone were located  $-8.9\pm1.3\%$  and  $+8.2\pm1.2\%$  from  ${\rm Fat}_{\rm max}$ . The  ${\rm Fat}_{\rm max}$  zone was found to be between  $55\pm3$  and  $72\pm4\%{\rm VO}_{2\rm max}$ , which is equivalent to  $68\pm3$  and  $79\pm3\%{\rm HR}_{\rm max}$ , respectively. At exercise intensities above the high border of the  ${\rm Fat}_{\rm max}$  zone, fat oxidation rates decreased markedly. The contribution of fat oxidation to energy expenditure became negligible above  $89\pm3\%{\rm VO}_{2\rm max}$  (range 71-99) and  $92\pm1\%{\rm HR}_{\rm max}$  (range 84-98).

The average total work done during the CE tests was  $2.79 \pm 0.03$  MJ. There were no significant differences in energy expenditure between the test days. Fat<sub>max</sub> was determined with the average fat oxidation rates of the CE tests. This exercise intensity was also determined with the fat oxidation rates measured during the first 5 min of each CE test. In Table 3, the exercise intensities at Fat<sub>max</sub> during GE, CE<sub>average</sub>, and CE<sub>5-min</sub> are presented. The exercise intensities are expressed as absolute and as percentage of maximal oxygen uptake and heart rate. There appeared to be no statistically significant differences in the Fat<sub>max</sub> determined with the three methods. The maximal fat oxidation rates measured during the three methods were also not statistically significant.

In Table 4, the "low" and "high" border of the  $Fat_{max}$  zone as well as  $Fat_{max}$  are expressed as a percentage of maximal oxygen uptake and maximal heart rate for the  $GE_{35/5}$ ,  $GE_{35/3}$  and  $GE_{20/3}$  exercise protocols. In addition,  $W_{max}$ ,  $\dot{V}O_{2max}$ , and  $HR_{max}$  reached during the three protocols are also shown in this table. No significant differences

TABLE 3. Exercise intensity at Fatmax

	Ÿ0,		Heart Rate	Fat Oxidation	
	(L·min <sup>−1</sup> )	%VO <sub>zmax</sub>	(bpm)	%HR <sub>max</sub>	(g·min−¹)
GE	2.48 ± 0.17	56 ± 3	132 ± 4	70 ± 2	$0.56 \pm 0.05$
CE <sub>average</sub>	$2.80 \pm 0.16$	64 ± 3	$135 \pm 3$	71 ± 2	$0.66 \pm 0.06$
CE <sub>5-min</sub>	$2.52 \pm 0.14$	58 ± 3	$125 \pm 3$	$66 \pm 2$	$0.69 \pm 0.06$

Values are mean  $\pm$  SEM: N = 18.

were found in maximal performance ( $W_{max}$ ,  $\dot{V}O_{2max}$ ) or  $HR_{max}$  between the three protocols. In Figure 2, the average fat oxidation curves for  $GE_{35/5}$ ,  $GE_{35/3}$ , and  $GE_{20/3}$  are presented. There were no significant differences in the fat oxidation rates between the three protocols. The exercise intensities demarking the  $Fat_{max}$  zone were also not significantly different between the three exercise tests.

#### DISCUSSION

The main goal of the present study was to develop an exercise protocol to determine the exercise intensity that elicits the maximum fat oxidation rate. Here we introduce the term Fat<sub>max</sub> for this exercise intensity at which maximal fat oxidation was observed. Although several studies reported maximal fat oxidation rates between 25 and 85% VO<sub>2max</sub> (28), no studies have systematically determined Fatmax. In this study, we have used an incremental exercise protocol with 5-min stages and 35-W increments to determine Fat<sub>max</sub>. Here Fat<sub>max</sub> was located at 64 ±  $4\% \dot{V}O_{2max}$ , corresponding to  $74 \pm 3\% HR_{max}$ . In addition to Fat<sub>max</sub>, a Fat<sub>max</sub> zone was also determined. This zone was defined as a range of exercise intensities with fat oxidation rates within 10% of fat oxidation rates at Fat<sub>max</sub>. The results of this study show that fat oxidation rates are within 10% of the peak rate over a relatively large range of intensities (between 55  $\pm$  3%  $\dot{V}O_{2max}$  and 72  $\pm$  4%  $\dot{V}O_{2max}$ : 68  $\pm$  $3\%HR_{max}$  and  $79 \pm 3\%HR_{max}$ ). It must be noted that the absolute rates of fat oxidation are dependent on carbohydrate intake. It has been shown in numerous studies that ingestion of carbohydrate in the hours before exercise reduces the rate of fat oxidation in a subsequent exercise bout (6,8,9,12,15,35). To prevent a CHO-induced decrease in fat oxidation rates, all exercise tests in this study were performed after a 10- to 12-h fast. However, although it is known that carbohydrate intake can influence the absolute rate of fat oxidation during exercise, it is not known whether the intensity at which this occurs is also influenced.

Romijn and colleagues (28) investigated substrate utilization at three exercise intensities (25, 65, and  $85\% \dot{V}O_{2max}$ ).

Five male subjects performed three exercise bouts on consecutive days. One major finding in their study was that fat oxidation increased from 25 to 65% and declined again at 85% VO<sub>2max</sub>, which is in agreement with the results of the present study. Bergman and Brooks (2) studied seven trained and seven untrained men exercising at 22, 40, 59, and 75% VO<sub>2max</sub> after an overnight fast. The fat oxidation rates in the trained individuals showed a peak at 40% VO<sub>2max</sub> (fat oxidation rates 0.25, 0.41, 0.31, and 0.09 g·min<sup>-1</sup>, respectively), whereas the untrained group reached peak oxidation values at 59% VO<sub>2max</sub> (0.12, 0.20, 0.27, and 0.06 g·min<sup>-1</sup>). Due to the limited number of exercise intensities in the above-mentioned studies, determination of Fat<sub>max</sub>, however, could not be very accurate.

It could be argued that Fat<sub>max</sub> does not only depend on a person's ability to oxidize fatty acids but also on the methods by which it is determined. By using a graded exercise protocol, Fat<sub>max</sub> can be influenced by a number of factors. A general problem with graded exercise tests is the fact that substrate utilization during the later stages of the test can be influenced by the previously performed exercise intensities. Another, maybe even more important factor, is the duration of the exercise performed at each exercise intensity, because the contribution of fat to energy production increases when exercise is continued for a long period (21,25,34). Here we studied the effects of previously performed exercise and exercise duration on the location of Fat<sub>max</sub>.

The results of the first 5-min of the CE tests have been used to investigate the influence of the previously performed exercise intensities. Fat<sub>max</sub> determined with this method was not significantly different from that found with the incremental test. It can therefore be concluded that the exercise intensities performed during the early stages of the incremental test do not change the exercise intensity at which fat oxidation rates are maximal. Similar results were found by Rieu et al. (26) in 1989. In this study, eight subjects performed a graded exercise test on a treadmill with 4-min stages. Subsequently, each subject performed exercise of 4-min duration, at the same speed as that used during the graded exercise test on separate days. It was reported

TABLE 4. Fat<sub>max</sub> zone as percentage of maximal performance.

	GE <sub>35/5</sub>	GE <sub>35/3</sub>	GE <sub>20/3</sub>			
W <sub>max</sub> (W) VO <sub>2max</sub> (L·min <sup></sup> )	385 ± 12 4.92 ± 0.13	389 ± 11 5.02 ± 0.1	357 ± 8 5.03 ± 0.08			
HR <sub>max</sub> (bpm)	185 ± 4		182 ± 4			
	%VO <sub>2max</sub>	%HR <sub>max</sub>	%VO <sub>2max</sub>	%HR <sub>max</sub>	%Ÿ0 <sub>2max</sub>	%HR <sub>max</sub>
Low	48 ± 3	64 ± 2	46 ± 3	62 ± 2	51 ± 2	65 ± 2
Fat <sub>max</sub>	$61 \pm 3$	$72 \pm 2$	59 ± 2	72 ± 1	65 ± 3	$74 \pm 3$
High	70 ± 3	78 ± 2	68 ± 4	77 ± 2	75 ± 4	$81 \pm 3$

Values are mean  $\pm$  SEM; N = 7.

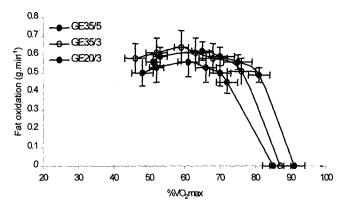


FIGURE 2—Fat oxidation rates versus exercise intensity for the  $GE_{35/5}$ ,  $GE_{35/5}$ , and  $GE_{20/3}$  tests; N=7; values are mean  $\pm$  SEM.

that oxygen uptake, carbon dioxide production, and ventilation rate were identical in the incremental exercise test and the isolated 4-min trials (26).

To study the effect of exercise duration, the CE tests were performed over a longer period of time. The exercise intensity at which fat oxidation peaked determined with this method was also not different than that found with the GE test. The maximal rate of fat oxidation was not statistically different between the GE,  $CE_{average}$ , or  $CE_{5-min}$  tests.

Recently, the relationship between percentages of maximal oxygen uptake or maximal heart rate and the individual anaerobic lactate threshold was investigated (23,32). In both studies, a large variation was found in the individual relationships between lactate concentrations and the tested intensities. It was concluded that the individual anaerobic threshold could not be predicted from fixed workloads in percentages of  $\dot{V}O_{2max}$  or  $HR_{max}$ . The same conclusions can be drawn for the prediction of  $Fat_{max}$ . The between-subject variation for both  $Fat_{max}$  and the rate of fat oxidation at  $Fat_{max}$  was large. Due to these large variations, it may not be possible to extrapolate  $Fat_{max}$  found in this group of subjects to individuals.

To reduce the influence of previously performed exercise intensities and the time required to perform the test, it would

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be advantageous if the duration of each stage could be reduced. It has been shown that submaximal work stages of 3-min result in reliable and valid measurements of  $\dot{V}O_2$  at LT (27,33). In addition, recently it has been shown that an exercise test with 3-min stages can be used to accurately determine  $\dot{V}O_{2max}$  (3). In accordance with these results, no differences in maximal performance (expressed as  $W_{max}$ ,  $\dot{V}O_{2max}$ , or  $HR_{max}$ ) were found between  $GE_{35/5}$  and  $GE_{35/3}$  protocols in the present study. The results of the present study also indicate that determination of  $Fat_{max}$  with an exercise protocol with 3-min stages will produce the same results as with a test with 5-min stages.

There could be a possibility that even 35-W increments are too large to accurately determine Fat<sub>max</sub>. Reducing the power increment to 20-W increases the number of measurements during a test. A practical disadvantage of the smaller increments would be the long duration of the exercise test. This study, however, showed that there are no differences in Fat<sub>max</sub> determined during a test with 35- or 20-W increments. The maximal performance achieved with this protocol was not significantly different from the maximal performance during the GE<sub>35/5</sub> and GE<sub>35/3</sub> protocols. Because 20-W increments did not seem to improve the detection of Fat<sub>max</sub>, for reasons of practicality, 35-W increments would be preferred.

In conclusion, the findings of the present study suggest that a continuous incremental exercise test on a cycle ergometer with 3-min stages and 35-W increments in work rate allows for valid assessment of Fat<sub>max</sub> and a Fat<sub>max</sub> zone in well-trained athletes. Fat oxidation rates appeared to be high over a large range of exercise intensities; however, at exercise intensities above Fat<sub>max</sub>, a marked drop in fat oxidation was observed.

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