Comparison between FAT Max and Maximal Fat Oxidation in Active and Sedentary Males

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Abstract

Background: The purpose of this study was to determine appropriate intensity of activity with FAT_max during incremental exercise in the active and sedentary male participants.

Material and methods: In this study, 11 active male students (VO₂max 42.87±1.75ml.kg⁻¹.min⁻¹, BMI 21.40±1.01 kg.m²) and 9 sedentary male students (VO₂max 36.57±2.95ml.kg⁻¹.min⁻¹, BMI 24.28±1.83kg.m²) were selected as the active and sedentary groups. Participants performed an incremental test with three minutes intervals on the treadmill. Exercise intensity was measured in all phases by measuring oxygen consumption. Also, heart rate and the fat oxidation was measured using indirect calorimetric. Independent t-test was used to compare the mean FAT_max in the two groups. Also two-way analysis of variance (ANOVA) with repeated measurements was used to compare FAT_max at 7 levels of exercise intensity between the two groups at α≤0.05 confidence interval level. Also, the Pearson correlation coefficient was used to measure the relationship between VO₂max and maximal fat oxidation (MFO).

Results: There was no significant difference between FAT_max of the active and sedentary groups, in terms of VO₂max and HR_max percentage, but the difference between MFO in the active and sedentary groups was significant (p=0.001). The results also showed that there are significant differences in fat oxidation during 7 levels of intensity training between the active and sedentary males (p=0.001). Also, there was a significant correlation between VO₂max and FAT_max of two groups (p=0.002).

Conclusion: Based on the results, it can be concluded that the active participants, due to their physiological adaptations with exercise, showed significant higher fat oxidation at FAT_max point and all phases of exercise intensity.

Keywords: maximal fat oxidation, FAT_max, exercise intensity, indirect calorimetry.

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**Introduction**

Fats and carbohydrates are the main sources of energy during the rest, exercise and training activities, they are also interrelated with factors such as duration and intensity of exercise, type of sport activity, physical fitness, the composition of food before the sport activities, environmental conditions and consumption of complementary foods. When intensity of an activity increases, the kind of fuel substance and its consumption change. The proportional share of carbohydrate oxidation in supplying energy increases progressively and thus the relative share of fat oxidation in supplying energy decreases, however, increase in intensity of an activity from low to moderate, increases the absolute value of fat oxidation (value of fat oxidation in grams per minute) and if the intensity increases, it gets decreased. In general, the highest level of fat oxidation has been reported to be at low intensity to moderate intensities. FATmax or the exercise intensity in which Maximal Fat Oxidation (MFO) occurs was introduced by Jeukendrup and Achten in 2001. Finding the effective factors on such a metabolic indicator, can lead us to a better understanding of it and helps us benefit from it appropriately in exercise and weight control or weight loss diets.

Some of the previous studies estimated the FATmax through incremental exercises on the ergometer with 2 to 4 intensity of training. Achten et al., and Achten and Jeukendrup, applying a multi-level graded exercise ergometer test, found that fat oxidation rate in exercise intensities between VO$_{2_{max}}$ %64±4 and 62±3 will reach its climax and then decreases dramatically in higher exercise intensities and reaches a slight level. A similar study on the ergometer by Nordby et al. showed that men who exercise had higher rates of MFO, and the rates were found in relatively higher FATmax intensities compared to untrained men (50 and 43% VO$_{2_{max}}$ respectively). In contrast, Stisen et al. concluded that there was no significant difference between the endurance in trained and untrained females in terms of FATmax on the cycle ergometer test. In fact, FATmax is equal among the trained and untrained individuals, and the difference is in terms of VO$_{2_{max}}$ and the enzyme activity of the participants. In other words, trained individuals have the MFO which occurs, due to higher VO$_{2_{max}}$, at a higher absolute intensity of activity and equal relative intensity of activity in comparison with untrained individuals.

Researchers have mainly studied the MFO only in 2 intensities of activity, or 4 exercise intensities, and the MFO is determined in only one of these intensity points. Therefore, it seems that this method in MFO evaluation might not have been precise. On the other hand, various studies on fat oxidation are conducted using an ergometer bike, and only a limited number of researches are done on treadmill, through incremental exercise, while according to Cheneviere et al.’s study the energy expenditure and FATmax during the exercise on ergometer bike and treadmill are different. Since major muscular mass are involved during an activity on treadmill compared to the ergometer. It is, therefore, possible to say that the greater storage and return of elastic energy may delay the onset of peripheral fatigue and/or reduce the recruitment of larger motor units which consist of more type II muscle fibers during running, in comparison to cycling for same relative exercise intensity. This may lead to higher CHO oxidation rates, higher lactate production and reduction in muscle PH in cycling, which can inhibit the activity of carnitine palmitoyltransferase I (i.e. key enzyme in fat metabolism). This may explain the lower Fatmax in cycling and the
rightward asymmetry in the running fat oxidation kinetics.

On the contrary, only a few researches have studied fat oxidation and FATmax rates across two groups of active and sedentary males in several intensities of incremental exercise on treadmill. To our knowledge only Stisen et al\textsuperscript{17} performed such a comparison among the trained and untrained women, but only on ergometer bike. Therefore, the purpose of this study was to quantitatively describe and compare MFO and FATmax and also compare the fat oxidation rates across different intensities during a graded exercise treadmill test, between the two groups of active and sedentary participants using an indirect calorimetry equation. It was hypothesized that both MFO and fat oxidation rates are different over a wide range of intensities in the active and sedentary participants.

**Material and methods**

**Participants**

11 active and 9 sedentary male students participated in this study approved by the South Birmingham Local Research Ethics Committee, UK. As assessed by a general health questionnaire, all the participants were healthy and none were accepted on to the study if diabetic or medically treated for heart or blood pressure irregularities. All Participants were informed on the purpose and nature of the study, after which their written, informed consent was gathered.

**General design**

To conduct this study, we selected 11 active male students (soccer players) and 9 sedentary male students who performed a graded exercise test to exhaustion level on a treadmill (PPS 70sport-I, Woodway, Weilam Rhein, Germany). Provided that their fat percentage was less than 30 and their BMI was less than 25 (to make the samples standard). The sedentary group had not participated in any regular sports activity during the last 3 years. In contrast, the active participants were students who had regular soccer training for at least 6 to 8 hours per week during the last 2 years. Individual characteristics of the participants are presented in table 2. Fat oxidation was determined by indirect calorimetry and plotted as a function of exercise intensity.

**Experimental design**

Before the experiments, the subjects were familiarized with the equipment and the procedures. Experiments were always performed in the morning (starting between 8 to 10 a.m) simultaneously to avoid circadian variance. Subjects were asked to fill in a 1-day food diary on the day before their first test. Subjects were reported to the laboratory after a 10-12h overnight fast. They had all been instructed to avoid strenuous exercise and alcohol for the preceding 24h. The participants’ weight and height were measured before eating breakfast and their fat was measured using bioelectric impedance method (Olympia 3.3). The exercise protocol used here was adapted from a previously described and validated protocol\textsuperscript{3,14} in which it was concluded that an incremental exercise test with stages of 3-min duration could be used to determine both MFO and FATmax. Running on treadmill (HP/Cusmose, Germany) began by speed of 3.5 km.h\textsuperscript{-1} and a gradient of 1%. Every 3 minutes, the speed increased by 1 km.h\textsuperscript{-1} until 6.5 or 7.5 km.h\textsuperscript{-1}. Then during all the 3 minute stages the speed was kept constant, but the gradient was increased by 2% to reach RER=1. Afterward, in the last stage up to the point of exhaustion, speed and gradient were increased every 2 minutes (2% gradient and 1 km.h\textsuperscript{-1} speed)\textsuperscript{2}. The aim of the final section of the exercise test was to obtain a measure of VO\textsubscript{2max} within a short time. VO2max was reached when: 1) RER>1.10, 2) level of oxygen consumption did not increase in spite of the increase in speed and
slope of treadmill and 3) maximum heart rate (220 pulse per minute-age) was provisioned to be 10 pulses per minute around the maximal zone\textsuperscript{13}. Breath-by-breath measurements were taken throughout exercise by using an automated gas-analysis system (GANSCHORN, Germany). The gas analyzers were calibrated with a 4.95% CO\textsubscript{2}-95.05% N\textsubscript{2} gas mixture (BOC Gases, Surrey, UK), and the volume transducer was calibrated with a 3-liter calibration syringe (GANSCHORN, Germany). Heart rate was recorded continuously by telemetry using a heart-rate monitor (Polar Vantage NV, Polar Electro Oy, Kempele, Finland).

**Indirect calorimetry and calculations**

Oxygen uptake (V\textsubscript{O}\textsubscript{2}) and carbon dioxide production (V\textsubscript{CO}\textsubscript{2}) were averaged over the last 2 minutes of each exercise stage, during which the RER was <1. For each of these stages, fat oxidation was calculated using indirect calorimetry equations,\textsuperscript{21} with the assumption that the amount of urinary nitrogen is negligible. Substrate oxidation rates were then plotted as a function of exercise intensity, expressed as a percentage of maximal oxygen uptake (VO\textsubscript{2max}) and percentage of maximal heart rate (HR\textsubscript{max}). From each fat oxidation curve, several features were identified 1) MFO, the peak rate of fat oxidation measured over the entire range of exercise intensities, and 2) FAT\textsubscript{max}, the exercise intensity at which the fat oxidation rate was maximal.

\[
\text{Fat oxidation (g.min}^{-1}) = 1.67 \times \text{VO}_{2} - 1.67 \times \text{VCO}_{2}
\]

In the above equation, VCO\textsubscript{2} and VO\textsubscript{2} are in liters per minute (L.min\textsuperscript{-1}) and fat oxidation is grams per minute (g.min\textsuperscript{-1}). The fat oxidation rate during each stage was measured by putting the amounts of VCO\textsubscript{2} and VO\textsubscript{2} in this equation and the appropriate intensity for it (FAT\textsubscript{max}) was measured in every stage according to a percent of VO\textsubscript{2max}. Firstly, by converting the amount of oxygen consumption from liters per minute unit (L.min\textsuperscript{-1}) to milliliter per minute (ml.min\textsuperscript{-1}) and then divided by VO\textsubscript{2max}. The exercise intensity was determined according to a percent of HR\textsubscript{max} by dividing the heart rate in every stage of the activities by maximal heart rate. The FAT\textsubscript{max} was recorded for each person according to a percent of VO\textsubscript{2max} and HR\textsubscript{max}.

**Statistical analysis**

For analysis of the MFO, FAT\textsubscript{ax} (VO\textsubscript{2max} %), and FAT\textsubscript{max} (HR\textsubscript{max}%) rate between active and sedentary individuals, the independent t-test was used, and for analysis of the 7 activity intensity stages, a one-way ANOVA 2 (group) × 7 (activity intensity) with a repeated measure on the last factor was performed. Upon observation of a significant F-test, post hoc LSD tests were conducted to identify the source of any significant main effects. In order to investigate the relationship between MFO and VO\textsubscript{2max}, Pearson's correlation coefficient was computed. Also, SPSS version 16 software was used for statistical analysis. The significant differences' level for all analyses was set at \( p \leq 0.05 \).

**Results**

Prior to research question's testing, an examination of the Kolmogrov Smirnov test of the data was undertaken to secure normal distribution assumption. The data generally was normally distributed(table 1). Individual characteristics of participants are presented in table 2 according to the group. The results of independent t-test revealed that there was significant difference between the active and sedentary groups in MFO (t(1.18)=4.43, \( p<0.05 \)), but, there was no significant difference between both groups in FAT\textsubscript{max} (VO\textsubscript{2max}% ) (t(1.18)=-0.66, \( p<0.05 \)) and FAT\textsubscript{max} (HR\textsubscript{max}% ) (t(1.18)=-1.27, \( p<0.05 \)) (See table 3, Figure 1 and 2). The data presented in Figure 1 shows that the MFO in the active group is 0.29±0.03 grams per minute and 0.23±0.02 grams in
the sedentary group; regarding the quantities of horizontal axis in the Figure for both active and sedentary groups, these results are respectively appropriate for an FATmax equal to 40.09±2.58 and 42.72±3.01% of VO2max. Also the quantities mentioned as MFO are appropriate respectively for intensities of activity equal to 56.45±4.33 and 60.09±3.37% of HRmax within the two active and sedentary groups (Figure 2). Prior to examination of the FATmax rate in activity intensities, the results of Mauchly’s test of Sphericity indicated that the error covariance matrix of dependent variable was the same.

For the fat oxidation rate in activity intensities, the results of a one-way ANOVA with repeated measure significant effects for groups F(1,18)=23.12, p=0.001, η2=0.167. An independent r-test analysis indicated that there was a difference between active and sedentary groups in stage 1 (t(1,18)= 3.66, p=0.002), stage 2 (t(1,18)=2.65, p=0.01), stage 3 (t(1,18)=2.98, p=0.008), stage 4 (t(1,18)= 3.54, p=0.002), stage 5 (t(1,18)= 2.16, p=0.045), and stage 7 (t(1,18)=2.38, p=0.028). The comparison of fat oxidation rate during stages indicated that the fat oxidation rate significantly superior in the active group than in the sedentary group, and the maximal fat oxidation rate occurred in stage 4 (see table 5, 6, and figure 3). The stages main effect was significant F(6,108)=46.21, p=0.001, η2=0.020, indicating that participants fat oxidation rate varied according to the 7 activity intensity stages, but groups by stages interaction effect F(6,108)=0.72, p=0.63, η2=0.091 was not significant (see table 5).

Post hoc Tukey test revealed that there was a significant difference between the active and the sedentary individuals (p<0.05), and the fat oxidation rate significantly was superior in the active individual (M=0.21, SD=0.01), than sedentary individual (M=0.17, SD=0.01) (see Figure 3). Pearson correlation coefficient indicated a statistically significant relationship between MFO and VO2max at the 0.05 level (r=0.64, p=0.002).

**Table 1. A sample Kolmogorov-Smirnov test**

<table>
<thead>
<tr>
<th></th>
<th>FATmax (%VO2max)</th>
<th>FATmax (%HRmax)</th>
<th>MFO</th>
<th>VO2max</th>
<th>Year</th>
<th>Height</th>
<th>Weight</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>Kolmogorov-Smirnov Z</td>
<td>0.53</td>
<td>0.41</td>
<td>0.59</td>
<td>0.47</td>
<td>1.22</td>
<td>0.72</td>
<td>0.64</td>
</tr>
<tr>
<td>Sig</td>
<td>0.93</td>
<td>0.99</td>
<td>0.87</td>
<td>0.97</td>
<td>0.10</td>
<td>0.67</td>
<td>0.80</td>
</tr>
</tbody>
</table>

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Table 2. Individual characteristics of participants.

<table>
<thead>
<tr>
<th>Feature</th>
<th>Active group (n=11)</th>
<th>Sedentary group (n=9)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>22.36±1.43</td>
<td>24±1.11</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>174.18±3.81</td>
<td>177.88±6.69</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>64.40±3.60</td>
<td>76.11±4.04</td>
</tr>
<tr>
<td>BMI (kg.m$^{-2}$)</td>
<td>21.40±1.01</td>
<td>24.28±1.83</td>
</tr>
<tr>
<td>$VO_{2max}$ (ml.kg$^{-1}$.min$^{-1}$)</td>
<td>42.85±1.75</td>
<td>36.58±2.95</td>
</tr>
<tr>
<td>LBM (kg)</td>
<td>53.89±2.43</td>
<td>59.70±3.57</td>
</tr>
<tr>
<td>PBF (%)</td>
<td>17.26±1.75</td>
<td>22±3.14</td>
</tr>
</tbody>
</table>

1. lean body mass
2. percentage body fat

Table 3. Comparison of average $FAT_{max}$ ($VO_{2max}$ %), $FAT_{max}$ ($HR_{max}$ %) and MFO between the two groups.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Active mean±s</th>
<th>Sedentary mean±s</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$FAT_{max}$ (%$VO_{2max}$)</td>
<td>40.09±2.58</td>
<td>42.72±3.01</td>
<td>0.515</td>
</tr>
<tr>
<td>$FAT_{max}$ (%$HR_{max}$)</td>
<td>56.45±4.33</td>
<td>60.09±3.37</td>
<td>0.353</td>
</tr>
<tr>
<td>MFO (g.min$^{-1}$)</td>
<td>0.29±0.03</td>
<td>0.23±0.02</td>
<td>0.001*</td>
</tr>
</tbody>
</table>

* Significant difference between groups ($\alpha < 0.05$)

Table 4. Mauchly's Test of Sphericity

<table>
<thead>
<tr>
<th>Within participants effect</th>
<th>Mauchly's W</th>
<th>Chi-Square</th>
<th>df</th>
<th>sig</th>
</tr>
</thead>
<tbody>
<tr>
<td>$FAT_{max}$</td>
<td>0.017</td>
<td>64.60</td>
<td>20</td>
<td>0.08</td>
</tr>
</tbody>
</table>

Table 5. Comparison of fat oxidation rate during stages between the two groups.

<table>
<thead>
<tr>
<th>Stages</th>
<th>Active mean±s</th>
<th>Sedentary mean±s</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.15±0.02</td>
<td>0.11±0.01</td>
<td>0.002*</td>
</tr>
<tr>
<td>2</td>
<td>0.18±0.03</td>
<td>0.15±0.02</td>
<td>0.01*</td>
</tr>
<tr>
<td>3</td>
<td>0.24±0.02</td>
<td>0.20±0.03</td>
<td>0.008*</td>
</tr>
<tr>
<td>4</td>
<td>0.23±0.02</td>
<td>0.23±0.02</td>
<td>0.002*</td>
</tr>
<tr>
<td>5</td>
<td>0.28±0.04</td>
<td>0.22±0.01</td>
<td>0.045*</td>
</tr>
<tr>
<td>6</td>
<td>0.20±0.02</td>
<td>0.17±0.03</td>
<td>0.056</td>
</tr>
<tr>
<td>7</td>
<td>0.17±0.03</td>
<td>0.13±0.04</td>
<td>0.028*</td>
</tr>
</tbody>
</table>

* Significant difference between groups ($\alpha < 0.05$)
Table 6. Analysis of variance for fat oxidation rate in activity intensities

<table>
<thead>
<tr>
<th>source</th>
<th>Type III sum of squares</th>
<th>df</th>
<th>Mean square</th>
<th>F</th>
<th>Sig</th>
</tr>
</thead>
<tbody>
<tr>
<td>stages</td>
<td>0.217</td>
<td>6</td>
<td>0.036</td>
<td>46.217</td>
<td>0.001</td>
</tr>
<tr>
<td>Stages* groups</td>
<td>0.003</td>
<td>6</td>
<td>0.001</td>
<td>0.721</td>
<td>0.634</td>
</tr>
<tr>
<td>Groups</td>
<td>0.052</td>
<td>1</td>
<td>0.052</td>
<td>23.123</td>
<td>0.001</td>
</tr>
<tr>
<td>Error (stages)</td>
<td>0.084</td>
<td>108</td>
<td>0.001</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Error (groups)</td>
<td>0.041</td>
<td>18</td>
<td>0.002</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 1. The amount of appropriate fat oxidation with intensity of activity according to % VO\(_{2}\text{max}\)

Figure 2. The amount of appropriate fat oxidation with intensity of activity according to % HR\(_{\text{max}}\)
Discussion

The aim of the present study was to find an intensity of sport activity, which has the highest productivity in fat metabolism for people. Clearly, exercise in intensities higher than the zone of FAT$_{\text{max}}$ causes fatigue or lactic acidity, the exercise in intensities lower than this level does not guarantee a necessary and sufficient benefit as an individual expects from exercise. Various studies have reported the MFO in the low to moderate intensities (between 33 to 65 percent VO$_{2\text{max}}$).\textsuperscript{7,10,13,22} However, most studies have used 2 to 4 intensity protocols to determine the MFO, and it seems that there might be more complete methods to estimate the MFO. We examined fat oxidation in an incremental exercise protocol including seven stages with seven different intensities of activity in order to determine, more precisely, the appropriate intensity of activity that elicits maximal fat oxidation, known as FAT$_{\text{max}}$. Furthermore, recent studies have examined only one sample population\textsuperscript{3,22}, while we studied two groups of active and sedentary participants, and the fat oxidation was examined at different intensities of activity across the two groups.

The results of this study showed that during an incremental exercise protocol, there was no significant difference between the active and sedentary groups in levels of FAT$_{\text{max}}$. The results of the study, therefore, supported the conclusions in studies by Lima-Silva et al.,\textsuperscript{23} Bogdanis et al.,\textsuperscript{24} Stisen et al.,\textsuperscript{17} and Achten and Jeukendrup,\textsuperscript{15} and they are found not compatible with the conclusions of studies by Bergman and Brooks,\textsuperscript{6} and Bircher and Knechtle.\textsuperscript{25} This incompatibility in Bergman and Brooks might stem from their exercise protocol, because they have examined fat oxidation in 4 intensities and as mentioned earlier, studies which examine fat oxidation in 2 to 4 intensities are
inaccurate and do not precisely show the \( \text{FAT}_{\text{max}} \) point, and they are likely to show \( \text{FAT}_{\text{max}} \) at a point between these 4 intensities. The reason why the conclusion by Bircher and Knechtle is rejected is that they used limited number of intensities, and also it can be due to their instrument (ergometer bike), because fat oxidation patterns on treadmill and ergometer bike are different, and this can be a justification for the inconsistency. In our study and the previous studies, there was no significant difference in levels of \( \text{FAT}_{\text{max}} \), between the active and sedentary groups with high and low \( \text{VO}_{2\text{max}} \) or in men and women. For example, in the study by Lima-Silva et al.,
 the \( \text{FAT}_{\text{max}} \) rate in the group with a low \( \text{VO}_{2\text{max}} \) was somehow higher than the same rate in the group with a high \( \text{VO}_{2\text{max}} \) (64.4±14.9% and 61.6±15.4 %\( \text{VO}_{2\text{max}} \), respectively). These differences might be due to interpersonal and inherent differences, in terms of reaching the MFO. On the other hand, occurrence of MFO in the active and sedentary group at equal intensities can be a result of high \( \text{VO}_{2\text{max}} \) level in the active group and their more enzyme activity. In other words, due to higher \( \text{VO}_{2\text{max}} \), the active individuals have the MFO, which occurs at a higher absolute intensity of activity in comparison with the sedentary individuals, and this intensity is relatively equal to the intensity of activity for the sedentary participants. That is, the MFO occurs at equal relative intensities and unequal absolute intensities. But in the present study probably due to the proximity between \( \text{VO}_{2\text{max}} \) level in the active and sedentary groups, \( \text{FAT}_{\text{max}} \) occurred in equal absolute and relative intensities (fourth stage).

The results of this study also showed that there is no significant difference between the active and sedentary groups in terms of the intensity of exercise according to a percent of \( \text{HR}_{\text{max}} \) which the highest fat oxidation rate occurs (\( \text{FAT}_{\text{max}} \)). This result confirms the findings of the study by Bogdanis et al.\(^{24} \) As mentioned earlier, we examined the intensity of activity based on two indexes of \( \text{VO}_{2\text{max}} \) and \( \text{HR}_{\text{max}} \). Regarding the percent of \( \text{VO}_{2\text{max}} \) and \( \text{HR}_{\text{max}} \), the relationship between intensity of activity was different in the active and the sedentary groups.\(^{26} \) In the present study, the MFO in the sedentary group occurred at 60\% \( \text{HR}_{\text{max}} \) (appropriate with 42\% \( \text{VO}_{2\text{max}} \)). This appropriateness is about 2 or 3\% higher than the active group. Such differences were interesting in the study by Venables et al.,\(^{2} \) which was conducted on a large number of samples. In their research 61\% \( \text{HR}_{\text{max}} \) was equal to 48\% \( \text{VO}_{2\text{max}} \). When the intensity of activity is according to a percent of \( \text{HR}_{\text{max}} \), these differences can be considered for estimating the desirable intensity of activity, using the heart rate.

Results show that levels of MFO in the active participants is significantly higher than in sedentary participants (0.29±0.03 and 0.23±0.02 g.min\(^{-1} \) for the active and the sedentary participants). These results are consistent with findings of studies by Lima Silva et al.,\(^{23} \) Bogdanis et al.,\(^{24} \) and Bergman and Brooks,\(^{6} \) however, they are inconsistent with the study by Stisen et al.\(^{17} \) The reason behind this inconsistency can be attributed to the issue that levels of \( \text{VO}_{2\text{max}} \) in the present study is similar to the studies by Achten et al.,\(^{3} \) and Achten and Jeukendrup,\(^{15} \) determined by the classified test from constant to fatigue (30 to 35 minutes), while in Stisen et al.’s study\(^{17} \), the \( \text{VO}_{2\text{max}} \) test was performed in a separate day, and when the participants were completely recovered, they used a shorter constant incremental test (6 to 9 minutes) up to an agreeable fatigue. Another reason for this inconsistency might have been due to the gender of participants. Because in Stisen et al.’s study\(^{17} \), the participants were trained and untrained females, while in the present study, the
participants were active and sedentary male students. As it is shown in studies by Venables et al., women have more \( \text{FAT}_{\text{max}} \) and MFO compared to men. Therefore, it might be safe to assume that the higher level of fat oxidation in women is a reason why there was no significant difference between participants of Stisen et al.’s study in terms of MFO.

In sarcoplasmic transport of fatty acids in various cells, many plasma membrane proteins are involved. These proteins in muscle cells include fatty acid binding protein plasma membrane (FABP\(_{pm}\)), fatty acid transporter (FAT/CD36) and fatty acid transports protein (FATP). These proteins increase due to physiological adaptations with exercise in active people. Major physiological adaptations to endurance exercise training (Such as increased capillary density), improves the distribution of oxygen (glucose and FFA) to the active muscles and increases the efficiency of trained muscles in the absorption of available oxygen. In comparison with inactivity condition, due to increased density of mitochondria, oxidative enzyme capacity of muscle fibers was twice as much. In addition to the increase in the enzyme activities of the electron transport chain, fat-oxidizing enzyme activity, including enzymes involved in fatty acid beta-oxidation are increased, as well. Furthermore, doing exercise helps lipoprotein lipase activity increase and thus, absorption circulating VLDL for oxidation in muscle becomes greater and the source of intramuscular triglyceride in trained individuals gets higher. During the exercise, plasma insulin level in trained individuals is higher than in untrained individuals, and also lactate production is lower in trained individuals, both changes further strengthen the lipolysis. After the exercise, consumption of intramuscular triglyceride sources might increase. Therefore, endurance training strengthens the fat oxidation through the active muscles.

In addition to these results, our study also revealed that there was a significant difference between the active and the sedentary groups in terms of fat oxidation level in seven stages of activity. These findings are not consistent with findings of Stisen et al.’s study. In their study, the significant difference in terms of fat oxidation was only reported to be at moderate and high intensities, while in our study, there was a significant difference in all the stages. In general, the active individuals significantly oxidize more fat than the sedentary ones, because they have more enzyme activity and higher fat oxidation capacity than the sedentary individuals. Thus in our study, in spite of less weight and lower BMI compared to sedentary participants, the active participants had higher capacity for oxidation of fatty acids, since they had gained more capacities through physical activities. However, the question is what causes the difference between various stages of this exercise protocol in terms of fat oxidation level in the active and the sedentary individuals? Increase in fat oxidation from rest to moderate intensities is often caused by the increase of access to FA. The rate of appearance for FA increases by an increase in lipolysis and a decrease in FA re-esterification. Wolfe et al. reported that the percent of re-esterification decreases from 70% in rest to 25% during 30 minutes of low to moderate intensities. This decrease in composition with the tripled increase in FA release from triacylglycerol (TAG) hydrolysis leads to an increase of FA access by 6 times for oxidation. In addition to more access to FA, transfer of FA from fat fiber toward active muscle increases, as well. However, when the intensity increases up to
high levels, there is no simultaneous increase in Glycerol RA.
Furthermore, the present study showed that there was a significant relation between the index of VO\textsubscript{2max} and MFO in the active and the sedentary participants. These findings confirm the findings of studies by Achten and Jeukendrup\textsuperscript{15} and Venables et al.\textsuperscript{2} and Stisen et al.\textsuperscript{17} The real association between the quantities of MFO and VO\textsubscript{2max} can be due to the physiological compatibilities acquired by exercise, which increase the aerobic capacity. Endurance training can significantly increase the fat oxidation during aerobic exercises increasing aerobic power. The information driven from cross-sectional studies by\textsuperscript{6,11,31,32,33} and linear studies by\textsuperscript{6,34,35,36,37,38} supports the idea that exercise decreases the reliance on carbohydrate (CHO) as a source of energy, and therefore increases the fat oxidation during aerobic exercise. Exercise researches mostly examined thin young men, but the observed increase of fat oxidation is not limited only to this specific group. Similar studies are done on women,\textsuperscript{18,38} elders\textsuperscript{34,36,39} and the obese.\textsuperscript{40} In other words, when doing an activity at an equal relative intensity, a person with higher VO\textsubscript{2max} can do more activity with higher absolute intensity, and consequently causing more energy consumption.

Finally, the results of the present study showed that the FAT\textsubscript{max} was similar in the active and the sedentary participants, and the only difference was in VO\textsubscript{2max} amounts of the participants. In other words, due to a higher VO\textsubscript{2max}, active individuals are likely to have the MFO which occurs at a higher absolute intensity (work load) of activity in comparison with the sedentary individuals. This intensity is relatively equal to the intensity of activity for the sedentary participants.

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**References**