Exercise Protocols to Estimate Fatmax and Maximal Fat Oxidation in Children

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Consensus on the exercise protocol used to measure Fatmax (exercise intensity corresponding to maximum fat oxidation (MFO)) in children has not been reached. The present study compared Fatmax estimated using the 3 min incremental cycling protocol (3-INC) and a protocol consisting of several 10 min constant work rate exercise bouts (10-CWR) in 26 prepubertal children. Group Fatmax values were the same for 3-INC and 10-CWR (55% VO2peak) and 95% limits of agreement (LoA) were ± 7% VO2peak. Group MFO values were similar between protocols, although 95% LoA were -94 to 113 mg·min⁻¹. While 3-INC provides a valid estimation of Fatmax compared with 10-CWR, caution should be exercised when estimating MFO in prepubertal children.

The exercise intensity that promotes the maximum fat oxidation (MFO) rate has been termed Fatmax (1) and has received a recent surge in interest in young people (4,11,21,22,27,41). These studies in young people have identified Fatmax using incremental protocols with exercise stages of 3 (27), 3.5 (41), 4 (4,22), 5 (21) and 6 (11) min in duration. In contrast, others have continued to use a more traditional approach of isolated exercise bouts lasting 6 (34) and 8–10 (23) min with standardized recovery periods to assess fat oxidation at different exercise intensities in children and adolescents.

The major advantage of using an incremental protocol with short stages is that fat oxidation can be estimated across a wide range of exercise intensities and in a single visit to the laboratory. Conversely, the use of longer duration exercise bouts can limit the estimation of fat oxidation to only three (23) or five (34) different exercise intensities, precluding a precise estimation of Fatmax. Furthermore, there is a trade-off between exercise stage duration and the number of exercise intensities; as stage duration increases, the number of exercise intensities may diminish (e.g., 11,21,27) to such an extent that the advantage of the incremental protocol may be lost. Therefore, an incremental protocol with 3 min stages may be the preferred combination of stage duration and number of stages to estimate Fatmax.

Studies using incremental protocols to estimate Fatmax in young people have adapted the 3 min incremental protocol originally validated by Achten et al. (1) in trained adult males. However, to our knowledge, the 3 min incremental protocol
has not been validated in children specifically. The primary issues with a 3 min incremental protocol are whether a physiological steady state is attained before the onset of the sampling period and whether there is a residual (carry-over) effect from stage to stage as the increments progress that influence subsequent fat oxidation estimations. Oxygen and carbon dioxide kinetics research suggests that children attain a steady state faster than adults and VO2 time constant values indicate the attainment of steady state within 2 min (15,39). However, the time constant may be longer for VCO2 (13,39) and, consequently, the attainment of a steady state may be delayed. Furthermore, VO2, but not VCO2, kinetics may become progressively slower at higher work rate steps during incremental exercise with 3 min stages (40). We are not aware of studies that have systematically examined the potential residual effect on fat oxidation during incremental exercise in children. Yet, in adults, it has been demonstrated that prior bouts of exercise may increase fat oxidation during subsequent exercise when compared with a single bout of prolonged exercise (18) and active warm-up may influence fat oxidation during a subsequent exercise bout possibly by increasing acetylcarnitine (19,26) and reducing blood and muscle lactate concentrations (10,28).

Research investigating the potential influence of sex on Fatmax in children appears to be limited to just one study (21) and the vast majority of studies have only included boys (e.g., 11,27). Consequently, there is a need for studies to assess Fatmax in girls and also examine any potential sex differences.

Evidently, there is a lack of consensus on the type of protocol that should be used to determine Fatmax in young people and inconsistencies in the methods used to determine Fatmax limits interstudy comparisons (e.g., 27,34). Considering the recent interest in Fatmax and potential clinical relevance of increasing fat oxidation (6,7), it is important to validate a protocol suitable for estimating Fatmax in children specifically. Therefore, the aim of the current study was to compare Fatmax estimated using an incremental protocol with 3 min stages (3-INC) with Fatmax estimated from several 10 min constant work rate (10-CWR) exercise bouts in prepubertal children. In addition, inclusion of girls and boys allowed an exploration of an independent sex effect.

**Methods**

**Participants**

After gaining approval from the University Ethical Advisory sub-Committee, 30 prepubertal children (15 boys and 15 girls) aged 8–10 y volunteered to participate in the study (26 were included in the final analyses). Written informed consent was obtained from the primary carer and the participants provided their “willingness to participate”. Participants were screened using a health history questionnaire. Exclusion criteria included: known congenital heart disease, musculoskeletal problems, uncontrolled exercise-induced asthma, diabetes and epilepsy.

**Anthropometry**

Anthropometric characteristics were assessed and recorded before exercise trials. Stature was measured using a stadiometer (Holtain, Holtain Limited, Dyfed, UK)
to the nearest 0.01 m. Body mass (BM) was measured using a beam balance scale (Seca Model 888, Hamburg, Germany) to the nearest 0.1 kg. Body mass index (BMI) was calculated as body mass (kg) divided by stature squared (m²). Skinfold thickness was determined from three different sites (triceps, subscapular and medial calf) on the right hand side of the body using a Harpenden skinfold caliper to the nearest 0.2 mm (Baty International, England). Each site was measured three times by the same investigator and the median value for each site was used to estimate percentage body fat (% BF) according to Slaughter et al. (33). Fat free mass (FFM) in kg was estimated using the following equation:

\[
FFM = BM \left(1 - (\frac{\%BF}{100})\right)
\]

Waist circumference was measured midway between the 10th rib and the iliac crest (24) using a Gulick tape measure (Creative Health Products, Plymouth, MI). With the assistance of a primary home-based carer (parent/guardian), participants provided a self-assessment of their physical maturation using secondary sexual characteristics (36).

Apparatus

All exercise tests were performed on an electromagnetically-braked cycle ergometer (Excalibur Sport, Lode, The Netherlands). Gas exchange was measured on a breath-by-breath basis and displayed on-line using a portable metabolic cart (K4 b², Cosmed, Rome, Italy) with a bidirectional 28 mm turbine flowmeter to measure expired air volume. The flowmeter was attached to a facemask (Hans Rudolf, Shawnee, USA) of an appropriate size with a dead space volume of 32 mL, which was fitted carefully to the face and checked for leaks before each test. Expired gas was sampled continuously from the flowmeter at a rate of 300 mL·min⁻¹. Gas calibration was performed according to the manufacturer’s recommendations before every test with the K4 b² software using well ventilated room air and a bottled gas mixture containing 5% CO₂, 16% O₂, balance N₂ (Scott Medical Products, Plumsteadville, PA). The flowmeter was calibrated using a bidirectional 3.0 L volume calibration syringe (Hans Rudolf, Shawnee, USA). Heart rate (HR) was monitored and recorded throughout exercise tests using short-range telemetry (Polar Vantage, Polar, Kempele, Finland). All calibration procedures were carried out before each experimental test.

Gas Exchange Analyses

Ventilatory variables were collected on a breath-by-breath basis and interpolated into 1 s intervals for all tests. Oxygen consumption (VO₂) and carbon dioxide production (VCO₂) values during the final min of each stage (3-INC) or bout (10-CWR) were used for data analyses. Breath-by-breath responses occasionally contain values that are clearly artifactual, which may result from swallowing or coughing (20). Therefore, individual VO₂ and VCO₂ values that were >3 standard deviations (SDs) of the mean were removed (20). Average values for VO₂ and VCO₂ from the final min of each stage or bout were then calculated and used for subsequent analyses.
Experimental Design

In this cross-sectional study, participants were asked to visit the laboratory on five separate occasions ~7 days apart.

Visit 1: Cycling Peak VO2 Measurement. The children were habituated to the laboratory environment, equipment and exercise protocols. In particular, the children practiced exercising on the cycle ergometer at 60 revs∙min⁻¹ while breathing through the facemask and were asked to pedal at 60 revs∙min⁻¹ for all exercise trials.

Subsequently, an incremental test was completed to volitional exhaustion for the measurement of peak VO2 (VO2peak). Participants were asked to avoid strenuous exercise and caffeine on the day of the trial and food intake 2 hr before testing. Following a 2 min warm-up of unloaded pedaling, the work rate increased by 8 or 10 W·min⁻¹ (work rate increment dependent on body size) with a maximum duration of 15 min. The seat height and pedal cranks were adjusted for each child and replicated during subsequent measurements using the cycle ergometer.

Maximal effort was considered to have been reached if the participants demonstrated a plateau in VO2, i.e., change in VO2 < 2.1 mL·kg⁻¹·min⁻¹ over the final successive stages of the test (37) or at least 2 of the following secondary criteria were achieved: heart rate 95% of age-predicted HRmax (220-age); (3) and a respiratory exchange ratio (RER) ≥ 1.05, in addition to the participant demonstrating clear subjective symptoms of fatigue (2). The highest recorded 30 s moving average VO2 (mL·min⁻¹) during the exercise test was recorded as VO2peak.

Visit 2, 3 and 4: Fatmax Exercise Trials. Participants reported to the laboratory at 08:00 following a 12 hr over-night fast. With the assistance of a primary home-based carer (parent/guardian), the children were asked to record their food and drink intake in the 24 hr period before visit 2 and replicate this before visits 3 and 4. Participants also refrained from strenuous exercise on the day before exercise testing. A healthy breakfast was provided on the completion of exercise.

Visit 2 (Incremental Exercise Test). Participants completed a submaximal incremental exercise protocol (3-INC) for the determination of Fatmax. The work rate began at 0 W and increased by 6 or 8 W every 3 min (work rate increment dependent on body size). The test was terminated when the RER was ~0.95 or the participant was exercising above 80% VO2peak. The average number of stages completed was 9 (range 8–11), which corresponded to a total exercise duration of 27 min.

Visit 3 and 4 (Constant Work Rate Exercise Bouts). Participants completed 6 × 10 min constant work rate exercise bouts (10-CWR) at exercise intensities corresponding to those in 3-INC, for which Fatmax had been identified previously. The bouts were performed in a randomly assigned counter-balanced order over the 2 visits (3 bouts per visit) and were each separated by a 15 min rest period. The completion of this protocol resulted in an exercise duration of 60 min spanning ~2 hr in total.

Visit 5: Repeat Measurement of Cycling Peak VO2. Participants performed a second VO2peak test (see visit 1) for the confirmation of VO2peak. The highest VO2peak value from either visit 1 or 5 for each child was used for data analyses. The VO2peak
determined during visit 5 was significantly higher than that determined during visit 1 for the group ($t_{(25)} = -3.35$, 90% CI: -5.2 to -1.7, effect size (ES) = 0.56).

**Indirect Calorimetry and Fatmax Calculations**

Fat oxidation rates were calculated using the following stoichiometric equation recommended by Frayn with the assumption that the urinary nitrogen excretion rate was negligible (16), as used by Achten et al. (1) in the validation study with adult participants:

\[
\text{Fat oxidation (mg min}^{-1}\text{)} = 1.67 \times \text{VO}_2 \text{ (mL x min}^{-1}\text{)} - 1.67 \times \text{VCO}_2 \text{ (mL x min}^{-1}\text{)}
\]

Indirect calorimetry does not provide a valid estimation of substrate oxidation for exercise intensities >80–85% VO$_2$max in adults (29) or during the nonsteady state (16). Data > 80% VO$_2$peak were, therefore, not used. Confirmation of a steady state was achieved by checking the slope of the linear regression line for VO2 and VCO2 plotted against time during the final 5 min of each exercise bout for 10-CWR. The exercise bout was not included in data analyses if a steady state could not be confirmed (arbitrary slope > 0.2). On average, nine exercise stages (3-INC) and five bouts (10-CWR) were included in data analyses for each child.

For each individual, the results from 3-INC and 10-CWR were used to construct a 2nd order polynomial curve of fat oxidation rate against exercise intensity, expressed as % VO$_2$peak. The curve was used to estimate Fatmax (%VO$_2$peak), maximal fat oxidation (MFO, mg·min$^{-1}$) and the 5% Fatmax zone (range of exercise intensities with fat oxidation rates within 5% of MFO); (1). The HR corresponding to Fatmax was calculated using the relationship between % VO$_2$peak and HR. The

![Fat oxidation vs. Exercise Intensity](image_url)

**Figure 1** — An example of a graph of fat oxidation rate (mg·min$^{-1}$) against exercise intensity (%VO$_2$peak) used to determine the Fatmax and MFO from 3-INC and 10-CWR
mean(SD) $R^2$ values for the polynomial curves of fat oxidation against % VO$_{2\text{peak}}$ were 0.80(0.18) for 3-INC and 0.81(0.21) for 10-CWR. An example of a graph and polynomial fit used to estimate Fatmax using both protocols for a participant is displayed in Figure 1.

A simple visual method was also employed to identify Fatmax and MFO (27) to confirm the results from the modeled data, where MFO was taken as the highest recorded fat oxidation rate and Fatmax was taken as the corresponding exercise intensity for each protocol.

Statistical Analyses

Statistical analyses were completed using SPSS software version 16.0 for Windows (SPSS Inc, Chicago, IL, USA). Shapiro-Wilk tests were used to confirm normal distribution and Levene’s tests were used to confirm homogeneity of variance. Separate $2 \times 2$ mixed measures analysis of variance (ANOVA) repeated for protocol were used to examine the data for Fatmax and MFO. Student’s independent $t$ tests were used to compare the girls’ and boys’ characteristics shown in Table 1. Values are expressed as mean(SD) with corresponding 90% confidence intervals (CI) around the mean difference (35) and effect sizes (ES) were calculated (30).

Limits of agreement (LoA) were used to compare 3-INC and 10-CWR at the individual level (8). Systematic error (SE), or bias, was determined by calculating the mean difference between 3-INC and 10-CWR and the random error (RE) was the SD of the paired differences, as outlined by Bland and Altman (8). The LoA were then calculated by determining a 95% limit above and below the mean difference (bias ± (1.96 × RE)). Student’s paired $t$ tests were used to examine the correlation between the residuals and the mean (proportional error check) and the absolute residuals and the mean (random error check). The 95% LoA for Fatmax

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Participant Characteristics</th>
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<tbody>
<tr>
<td></td>
<td>Girls</td>
</tr>
<tr>
<td>Age (y)</td>
<td>9.3(0.6)</td>
</tr>
<tr>
<td>Body mass (kg)</td>
<td>35.1(6.1)</td>
</tr>
<tr>
<td>Stature (m)</td>
<td>1.37(0.06)</td>
</tr>
<tr>
<td>BMI (kg·m$^{-2}$)*</td>
<td>18.6(2.6)</td>
</tr>
<tr>
<td>Body fat (%)*</td>
<td>21.7(4.7)</td>
</tr>
<tr>
<td>FFM (kg)</td>
<td>27.2(3.5)</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>60.5(7.8)</td>
</tr>
<tr>
<td>Physical maturation†</td>
<td>1(0.5)</td>
</tr>
<tr>
<td>VO$_{2\text{peak}}$ (mL·kg$^{-1}$·min$^{-1}$)*</td>
<td>42(6)</td>
</tr>
</tbody>
</table>

Physical maturation is secondary sexual characteristics (36)
†median (interquartile range)
*difference between girls and boys
were compared with the estimated values for the 5% Fatmax zone (range of exercise intensities with fat oxidation rates within 5% of MFO) to determine their practical importance.

**Results**

**Participant Characteristics**

Complete data for 26 children (13 girls and 13 boys) were available for analyses. The four children excluded had \( R^2 \) values below the arbitrarily chosen threshold of 0.5 for the polynomial models or less than four 10 min CWR bouts available for analyses. The boys had a lower BMI (\( t(24) = 2.44, 90\% \text{ CI: } 0.61 \text{ to } 3.49, \text{ ES } = 0.45 \)) and \% BF (\( t(24) = 3.29, 90\% \text{ CI: } 2.82 \text{ to } 8.96, \text{ ES } = 0.56 \)) compared with the girls. The physical characteristics of the participants are displayed in Table 1.

**Peak Exercise Responses**

Peak VO\(_2\) values (Table 1) were higher in the boys compared with the girls (\( t(24) = -3.44, 90\% \text{ CI: } -13.9 \text{ to } -4.7, \text{ ES } = 0.57 \)). All participants included in the analysis achieved the criteria for the attainment of maximal effort. The mean(\( SD \)) peak responses for secondary criteria were RER 1.07(0.07) and HR 201(7) beats·min\(^{-1}\) or 96(4)% age-predicted HR\(_\text{max}\) (220-age).

**Fatmax**

**Group Comparison.** At the group level, Fatmax (%VO\(_2\)\(_\text{peak} \)) was the same for 3-INC and 10-CWR (\( F(1,24) = 0.0, 90\% \text{ CI: } -1.3 \text{ to } 1.3, \text{ ES } = 0 \)). The sex by protocol interaction (\( F(1,24) = 0.51, \text{ ES } = 0.14 \)) indicated that the small between protocol effect was independent of sex. Furthermore, the main effect for sex (\( F(1,24) = 1.32, \text{ CI: } -8.0 \text{ to } 1.6, \text{ ES } = 0.23 \)) showed that small differences in Fatmax between the girls and boys were not meaningful (Table 2).

<table>
<thead>
<tr>
<th>Table 2</th>
<th>Group Comparisons of Fatmax and MFO for 3-INC and 10-CWR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Girls</td>
</tr>
<tr>
<td>Fatmax % VO(<em>2)(</em>\text{peak} )</td>
<td>54(6)</td>
</tr>
<tr>
<td>Fatmax HR (beats·min(^{-1}))</td>
<td>141(15)</td>
</tr>
<tr>
<td>Fatmax %HR(_\text{max} )</td>
<td>71(7)</td>
</tr>
<tr>
<td>MFO (mg·min(^{-1}))</td>
<td>255(45)</td>
</tr>
<tr>
<td>MFO (mg·kg FFM(^{-1})·min(^{-1}))</td>
<td>9.4(1.4)</td>
</tr>
</tbody>
</table>
**Individual Comparison.** Individual paired data provided a systematic bias ± random error of 0 ± 4% VO2\text{peak}, resulting in 95% limits of agreement (LoA) of ± 7% VO2\text{peak}. Furthermore, 18 of the 26 participants had paired Fatmax values that were within 3% VO2\text{peak} of each other when comparing 3-INC and 10-CWR (Figure 2). Proportional bias was not evident from examination of the residuals and random errors were homoscedastic from examination of the absolute residuals (Table 3).

The 5% Fatmax zone spanned 45(6) to 65(9)% VO2\text{peak} for 3-INC and 47(6) to 63(9)% VO2\text{peak} for 10-CWR. Therefore, the 95% LoA were within the 5% Fatmax zone, suggesting that 3-INC provides a practically useful surrogate measure of 10-CWR (Figure 3). The limits of the 5% Fatmax zone also suggest fat oxidation rates remain high over a wide range of intensities.

**Table 3** Bias and Limits of Agreement for Fatmax and MFO Estimated Using 3-INC and 10-CWR

<table>
<thead>
<tr>
<th></th>
<th>Fatmax (%VO2\text{peak})</th>
<th>MFO (mg·min(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bias ± RE</td>
<td>0 ± 4</td>
<td>9 ± 53</td>
</tr>
<tr>
<td>95% LoA</td>
<td>-7 to +7</td>
<td>-94 to 113</td>
</tr>
<tr>
<td>Residual check</td>
<td>R value</td>
<td>0.10</td>
</tr>
<tr>
<td></td>
<td>90% CI</td>
<td>0.12</td>
</tr>
<tr>
<td>Absolute residual check</td>
<td>R value</td>
<td>-0.19</td>
</tr>
<tr>
<td></td>
<td>90% CI</td>
<td>-0.08</td>
</tr>
</tbody>
</table>

RE—random error; LoA—limits of agreement; CI—confidence interval

**Figure 2** — Bland-Altman plot of Fatmax (%VO2\text{peak}) for 3-INC and 10-CWR
MFO

**Group Comparison.** Group comparisons revealed similar MFO for 3-INC and 10-CWR when expressed as mg·min⁻¹ (F(1,24) = 0.83, 90% CI: -27 to 8, ES = 0.18) and mg·FFM⁻¹·min⁻¹ (F(1,24) = 1.2, 90% CI: -1.1 to 0.2, ES = 0.21). The sex by protocol interactions for both absolute (F(1,24) = 1.3, ES = 0.22) and scaled MFO (F(1,24) = 1.0, ES = 0.20) again indicated that the small between protocol effects were independent of sex. In addition, the main effect for sex for absolute (F(1,24) = 2.36, 90% CI: -46 to 2, ES = 0.30) and scaled MFO (F(1,24) = 1.99, 90% CI: -2.0 to 0.2, ES = 0.28) showed that again differences between the girls and boys were not meaningful (Table 2).

**Individual Comparison.** For the absolute MFO, the systematic bias ± random error were 9 ± 53 mg·min⁻¹ and subsequently 95% LoA were -94 to 113 mg·min⁻¹, showing considerable individual variability when comparing 3-INC and 10-CWR (Figure 4). Proportional bias was not evident from examination of the residuals and random errors were homoscedastic from examination of the absolute residuals (Table 3).

**Visual Analyses**

Visual analyses were consistent with the results from the polynomial modeled data. Fatmax values for 3-INC (55(10) % VO₂peak) and 10-CWR (56(8) % VO₂peak) were similar on a group basis (F(1,24) = 0.87, 90% CI: -2.7 to 0.8, ES = 0.19). Individual analysis showed that systematic bias ± random error was 1 ± 5, resulting in 95%

![Figure 3 — Visual representation of Fatmax limits of agreement fitting within the 5% Fatmax zone using group average Fatmax values](image-url)
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LoA of -9 to +11% VO2peak for Fatmax. Absolute MFO values for 3-INC (271(47) mg·min⁻¹) and 10-CWR (277(40) mg·min⁻¹) were similar again on a group basis (F(1,24) = 0.40, 90% CI: -22.8 to 10.5, ES = 0.13). Systematic bias ± random error was 6 ± 53, resulting in 95% LoA of -97 to +109 mg·min⁻¹ for absolute MFO.

Discussion

To our knowledge, we have demonstrated for the first time systematically that a 3 min incremental exercise protocol provides a valid estimation of Fatmax in prepubertal children compared with more prolonged isolated exercise bouts. Although the majority of studies have used incremental protocols to estimate Fatmax in children (e.g., 11,27), there are two primary issues with a 3 min incremental protocol (1): whether a physiological steady state is attained before the onset of the sampling period; and (2) whether there is a residual effect from stage to stage as the increments progress that influence subsequent fat oxidation estimations. Therefore, 10-CWR was selected to provide a valid representation of fat oxidation rates using indirect calorimetry (32) and Fatmax for each individual was compared for 3-INC and 10-CWR.

Fatmax for the group was identical when comparing the two protocols and individual analysis revealed that the 95% LoA were ± 7% VO2peak (Figure 2). These limits are small enough to recommend the use of 3-INC for the estimation of Fatmax for various reasons. Firstly, fat oxidation rates were within 5% of MFO (mg·min⁻¹) from 45 to 65% VO2peak (3-INC) and 47 to 63% VO2peak (10-CWR), thus the 95% LoA were within the limits of the 5% Fatmax zone (Figure 3). Furthermore, fat oxidation rates remained high (within 5% of MFO) over a wide range of exercise intensities, thus small under- or over-estimations of Fatmax are likely to have negligible affects on absolute fat oxidation rates. A strength of 3-INC is

Figure 4 — Bland-Altman plot of MFO (mg·min⁻¹) for 3-INC and 10-CWR
the estimation of fat oxidation at around ten different exercise intensities using a single test lasting approximately 30 min. The 2nd order polynomial R^2 values (0.80 for 3-INC, 0.81 for 10-CWR) indicate a moderate to good goodness of fit for both protocols. However, we were not able to estimate fat oxidation over the same number of exercise intensities for 3-INC and 10-CWR, as pilot work indicated a residual effect of over three repeated 10 min bouts on fat oxidation in a single fasted session similar to the up-regulation reported during prolonged steady state exercise (14). In addition, 3-INC is undoubtedly practically advantageous when compared with 10-CWR, with the latter requiring multiple visits to the laboratory and the replication of food intake and physical activity in the days preceding each measurement period.

The findings in the current study are in agreement with Achten et al. (1), where it was reported that an incremental protocol with 3 min stages can be used to identify Fatmax in trained adult males compared with longer isolated bouts. A more recent study comparing Fatmax using two incremental protocols in sedentary individuals reported an average underestimation of 2 W when using 3 compared with 6 min stages and a maximum difference of 8 W (9). Given the small magnitude of these between protocol differences, the practical implications are questionable. Therefore, our results support previous findings in adults that suggest only small differences are evident when comparing short and long exercise stages to estimate Fatmax (1,9). Furthermore, the VO2 and VCO2 kinetic response to moderate intensity exercise is faster in children compared with adults (15,39), supporting the use of short 3 min stages in children, although fat oxidation rates were not estimated in these studies.

Fatmax for the group occurred at 55% VO2_peak (3-INC and 10-CWR), corresponding to heart rate values of 141 (3-INC) and 137 (10-CWR) beats∙min^-1. Similar values for Fatmax have been reported in other studies of prepubertal children, with values ranging from 49 to 56% VO2_peak (11,27,41). The wide limits of the 5% Fatmax zone suggests that fat oxidation rates were high over a large range of exercise intensities, a finding that is also in agreement with Achten et al. (1). Therefore, prescribing exercise within the Fatmax zone rather than Fatmax specifically may be sufficient to promote high fat oxidation rates. However, individual prescription is required for exercise at Fatmax (or within the Fatmax zone) due to the large interindividual variability observed in the current study (40–73% VO2_peak) and other studies with adults (25). The 3 min incremental protocol provides a practical method for providing this individual exercise prescription. The large interindividual variation also suggests that fat oxidation should be assessed at exercise intensities as high as 73% VO2_peak in some individuals (Fatmax occurred above 60% VO2_peak in 6 out of the 26 participants), although previous studies have only identified fat oxidation rates up to 60% VO2_peak (11,41).

The findings related to MFO in the current study are less clear. On a group level, MFO was similar for 3-INC and 10-CWR (260 vs. 270 mg∙min^-1, respectively). However, the large 95% LoA (-94 to 113 mg∙min^-1) demonstrate considerable intra-individual variability when comparing 3-INC and 10-CWR (Figure 4). Achten et al. (1) also reported similar group values for fat oxidation rates, but the correlation data they provided did not allow this insight at the individual level. These results suggest that 3-INC may not provide a valid indication of MFO. However, the
exercise protocol may only be partially responsible for these differences in MFO as the residuals between 3-INC and 10-CWR were randomly distributed above and below the small bias. Although participants were asked to consume the same diet and to refrain from strenuous exercise on the day preceding each Fatmax exercise test, this may not have been adequate to control for inherent day to day variations in RER and fat oxidation (5) and even controlling food intake 36 hr before trials may not be sufficient (25). Initial muscle glycogen content and dietary fat intake are both determinants of resting and exercise metabolism; therefore, variations in either or both may have influenced our results (12,17).

A further finding of the current study was that sex did not influence Fatmax or MFO in prepubertal children. Higher absolute fat oxidation rates have been reported in obese pubertal boys compared with girls (21). These differences may result from puberty, which has been shown to influence Fatmax and fat oxidation (27). However, differences in VO2_peak, BMI and % body fat between the boys and girls may have affected the between sex comparison in the current study. Further research is required to examine the effect of sex on fat oxidation during exercise in young people when these factors have been carefully matched.

Possible limitations of the current study include the use of a 5% Fatmax zone to interpret the LoA rather than a previously defined clinical anchor. However, it is a reasonable assumption that a 5% reduction in MFO will have a small affect on total fat oxidation and thus the potential health benefits of exercising at MFO will continue to be promoted. Although steps were taken to increase the validity of indirect calorimetry for fat oxidation estimations (e.g., excluding data >80% VO2_peak, checking for a steady state in VO2 and VCO2), we assumed that the urinary nitrogen excretion rate was negligible and did not account for an increase in nonrespiratory carbon dioxide excretion that may have resulted in an underestimation of fat oxidation at some of the higher exercise intensities (31). Estimations of FFM were based on % BF values from skinfold measurements, which could introduce a source of error in MFO values expressed relative to FFM. It should also be acknowledged that fat oxidation increases with exercise duration in children (14), thus the fat oxidation values reported using short duration exercise stages are likely to underestimate fat oxidation during prolonged exercise. Finally, the current study only included healthy prepubertal children. Consequently, it is not possible to recommend the use of 3-INC for children with other conditions that may slow VO2 or VCO2 kinetics. Slower VCO2 kinetics in obese compared with nonobese children (13) suggests that slightly longer stages (~4 min) may be preferred for obese children (4,22). However, research in this area is inconclusive (13,38), thus future research investigating VO2 and VCO2 kinetics and the validity of 3-INC in obese children is warranted.

In conclusion, an incremental exercise test with 3 min stages provided a similar estimation of Fatmax compared with several 10 min constant work rate exercise bouts in prepubertal children. The 3 min incremental protocol is, therefore, recommended to provide an estimation of Fatmax using a wide range of intensities and for practical reasons. The estimation of Fatmax using a practical protocol should ensure optimal exercise prescription for maximizing fat oxidation during exercise that may help to manage obesity and other health-related conditions, such as type 2 diabetes.
References