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### **Assessing differences in cardio-respiratory fitness with respect to maturity status in highly trained youth soccer players**

Doncaster, Greg; Iga, John; Unnithan, Viswanath

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# 1 **Assessing differences in cardio-respiratory fitness with respect to** 2 **maturity status in highly trained youth soccer players**

## 3 **Abstract**

4 **Purpose:** Examine differences in measures of cardio-respiratory fitness and determinants of  
5 running economy with respect to maturity status, in a group of highly trained youth soccer  
6 players.

7 **Methods:** Twenty-one highly trained youth soccer players participated. On separate visits,  
8 players' peak oxygen uptake, running economy, at three different speeds (8km·h<sup>-1</sup>, 80%GET  
9 and 95%GET), and pulmonary oxygen uptake kinetics (VO<sub>2</sub> kinetics) were determined. Players  
10 also performed a Yo-Yo Intermittent Recovery test level 1 (Yo-Yo IR1). Players were  
11 categorised as either 'pre-PHV' or 'mid-PHV' using the measure of maturity offset.  
12 Independent *t*-tests and Cohen's *d* effect sizes were then used to assess differences between  
13 groups.

14 **Results:** The mid-PHV group were significantly taller, heavier and advanced in maturity status.  
15 Absolute measures of VO<sub>2peak</sub> were greater in the mid-PHV group, however, when expressed  
16 relative to body mass, FFM and theoretically derived exponents, VO<sub>2peak</sub> values were similar  
17 between groups. Pre-PHV group presented a significantly reduced VO<sub>2</sub> response, during  
18 relative submaximal running speeds, when theoretically derived exponents were used, or  
19 expressed as %VO<sub>2peak</sub>. VO<sub>2</sub> kinetics (*tau*) were faster during a low (standing) to moderate (95%  
20 GET) transition in the pre-PHV group. Yo-Yo IR1 performance was similar between groups.

21 **Conclusion:** While measures of VO<sub>2peak</sub> and Yo-Yo IR1 performance are shown to be similar  
22 between groups, those classed as Pre-PHV display a superior running economy at relative  
23 submaximal running speeds and faster *taus* during a low-moderate exercise transition, than  
24 their more mature counterparts.

## 25 **Introduction**

26 The growing professionalism of sport has resulted in an advancement in the application of early  
27 specialization and systematic training of talented young athletes, with the aim of achieving elite  
28 professional status (26). Consequently, a large emphasis is placed on the talent identification  
29 and talent development of young soccer players (38). This has resulted in research which aims  
30 to identify, examine and analyse the key physical and physiological characteristics of elite  
31 youth soccer players that are associated with superior soccer performance (38). However,  
32 confounding variables of growth and maturation complicate the analysis and interpretation of  
33 these measures within youth populations as well as the provision of coaching and training  
34 prescription toward these diverse populations. Indeed, both physiological and anthropometric  
35 changes can occur during adolescence, particularly during rapid periods of growth and  
36 maturation, these changes then impact upon physical performance and confound the issues of  
37 talent development and identification (17).

38

39 Currently, there is a plethora of research examining the impact of maturational differences in  
40 anaerobic fitness characteristics (speed, acceleration, jump power, etc.) (12, 17), highlighting  
41 the physical dominance of players who are at an advanced stage of maturity (26, 12). Although  
42 youth soccer players will compete within their respective age groups (same chronological age),  
43 the impact of growth and maturation can result in large variations in skeletal age within male  
44 youth soccer players aged between 10-18 years old. Subsequently, those individuals which are  
45 of an advanced maturity status will often appear as the taller, heavier and more physically  
46 developed individuals. This can often result in a selection bias towards players of an advanced  
47 maturity status, due to their superior levels of speed, power and strength (26, 27), leading to  
48 talented, but physically less mature players not attaining their full potential and dropping out  
49 of the game (12).

50 Research has highlighted the dominance of early maturing individuals in anaerobic fitness  
51 characteristics (27). The energy provision required for competitive soccer performance,  
52 however, is predominantly derived from aerobic energy sources (19). While an improved  
53 ability to activate anaerobic energy resources will result in superior performance in one-off  
54 explosive actions during match-play (sprints), an enhanced ability to utilise aerobic energy  
55 resources will (theoretically) result in an improved tolerance to fatigue and an ability to  
56 maintain levels of high intensity activities, and therefore maintain physical performance for a  
57 prolonged period of time (33). Therefore, while the less mature player may be unable to  
58 metabolically activate anaerobic energy sources, they will however, have the capacity to  
59 sustain high intensity activities via primarily aerobic energy pathways (33). Yet, there is little  
60 information regarding the impact of maturation with respect to detailed measures of cardio-  
61 respiratory fitness, in highly trained youth soccer players. Moreover, recent research has  
62 highlighted the importance of appropriate scaling to successfully accommodate the nonlinear  
63 relationship between body size descriptors and  $VO_{2peak}$  (10, 25, 1). Adopting the traditional  
64 ratio-standard method ( $mL \cdot kg^{-1} \cdot min^{-1}$ ) results in an adjusted  $VO_{2peak}$  which is not independent  
65 of body size, therefore research advocates the use of allometric scaling, when interpreting  $VO_2$   
66 data, to remove the confounding influence of body size, in individuals at different stages of  
67 maturation (10).

68

69 Therefore, the purpose of this study was to examine inter-group differences in ratio-standard  
70 and allometrically-adjusted measures of submaximal and peak cardio-respiratory fitness and  
71 determinants of running economy with respect to maturity status, in a group of highly trained  
72 youth soccer players. It was hypothesized that, when appropriately scaled, measures of  $VO_{2peak}$   
73 would be unaffected by maturity status. Players of a lower maturity status, however, would  
74 present a reduced  $VO_2$  response during all submaximal running speeds, when appropriately

75 scaled. Similarly, it was hypothesised that less mature players would display superior  $\text{VO}_2$   
76 kinetics (faster *taus*), than their more mature counterparts.

77

## 78 **Methods**

### 79 *Participants and Anthropometry*

80 Twenty one highly trained youth soccer players aged between 12 and 14 years volunteered to  
81 participate in this study. All participants were outfield players from the same Category One  
82 Premier League Football Academy. Prior to the commencement of the study, all players  
83 completed medical health questionnaires and training history questionnaires. Table 1 displays  
84 all anthropometric and screening measures. Maturity status was quantified using self-  
85 assessment Tanner Stage method (30, 36) and maturity offset (31). Sum of 4 skinfolds was  
86 assessed using Harpenden skinfold calipers (Cranlea, Birmingham, UK) to measure skinfold  
87 thickness at the biceps, triceps, subscapular and superilliac according to the procedures outlined  
88 by ISAK (28). Percent body fat was then estimated using the equation of Siri (35), this  
89 information was then subsequently used to estimate individuals' fat free mass (FFM). Players  
90 and their parents were informed about all procedures and requirements involved in the study  
91 before opting to provide written informed consent and assent from parents and participants,  
92 respectively. Ethical approval was granted from the local university ethics committee.

93

### 94 *Calculation of Maturity Offset*

95 Players were categorised into pre-PHV and mid-PHV groups using the calculation of maturity  
96 offset (31). Measurements obtained to gain an estimate of each individual's maturity offset  
97 included stretch standing stature and stretch sitting stature, to the nearest mm (Seca 217,  
98 Birmingham, United Kingdom) as well as body mass, to the nearest 0.1 kg (Seca 761,  
99 Birmingham, United Kingdom). As described by Mirwald et al. (31) two measurements were

100 taken for each anthropometric measure, with a third required if the first two measures differed  
101 by more than 4 mm for measures of stature and 0.4 kg for weight. For each anthropometric  
102 measure these were then averaged. All measurements were obtained by a sport practitioner  
103 experienced in the assessment of such procedures. This data was then inputted into a specially  
104 designed Microsoft Excel Spreadsheet which employed the maturity offset equation of  
105 Mirwald et al. (31) to provide an estimated value for the years from peak height velocity (PHV)  
106 for each individual.

107

108 Once the maturity offset was known, participants were classified as either pre-PHV or mid-  
109 PHV. Players who were  $\geq -4.0$  and  $< -1.0$  year away from their respective period of PHV were  
110 classified as pre-PHV ( $n = 10$ ), while those who were between  $\geq -1.0$  and  $< +0.99$  years from  
111 their predicted PHV were classified as mid-PHV ( $n = 11$ ). The maturity offset measure gives  
112 an indication to an individual's maturity status, as such, those who were classified as mid-PHV  
113 were at an advanced stage of maturation, when compared to their counterparts who were  
114 classified as pre-PHV.

115

116 **Table 1:** Anthropometric and screening measures ( $n = 21$ )

117 \*\*\*Insert Table 1 Here\*\*\*

118

### 119 *Study Design*

120 Players' anthropometric measurements were collected first, followed by laboratory then field  
121 tests. The players visited the laboratory on 4 separate occasions. During the first visit, players  
122 performed an incremental, ramp treadmill protocol for the assessment of players' gaseous  
123 exchange threshold (GET),  $VO_{2peak}$  and the speed corresponding to 60% of the difference  
124 between GET and  $VO_{2peak}$  ( $60\% \Delta$ ). Players then returned to the laboratory for a second time,

125 following a minimum of 24 hours of recovery. On the second visit, players were required to  
126 run for 4 min, at progressive sub-maximal intensities ( $8\text{km}\cdot\text{h}^{-1}$ , 80%GET and 95%GET), with  
127 2 min passive recovery between speed increments. Players returned to the laboratory on two  
128 further occasions, each following a minimum of 24 hours of recovery. These two visits to the  
129 laboratory were identical and required the participant to complete a work-to-work protocol (rest  
130 - 95%GET – 60% $\Delta$ ) on a motorised treadmill for the assessment of their pulmonary oxygen  
131 uptake kinetics ( $\text{VO}_2$  kinetics). Following all laboratory tests and a minimum of 48 hours  
132 recovery, players completed a maximal Yo-Yo Intermittent Recovery test level 1 (Yo-Yo IR1)  
133 (6). For each participant, all testing was completed at the same time of day ( $\pm 2$  hours), with  
134 the ranges for room temperature, humidity and pressure corresponding to  $18.7 - 22.8$  °C, 62 -  
135 63 % and 1011 - 1021 mmHg respectively, for laboratory testing.

136

### 137 *Assessment of Peak Oxygen Uptake and Gaseous Exchange Threshold*

138 Upon arrival to the laboratory and following the necessary screening procedures, participants  
139 were fitted with a Polar Heart rate monitor (Polar Electro, Kempele, Finland) and face-mask  
140 (Hans Rudolph, Hans Rudolph, Kansas City, USA), which was connected to an online gas  
141 analysis system (Cortex MetaMax 3B, Cortex Biophysik GmbH, Leipzig, Germany). The  
142 online gas analyser was calibrated prior to each visit according to the manufacturer's  
143 instructions, using a known gas concentration and a 3-L syringe for manual volume calibration  
144 of the ventilation sensors. Prior to each test, participants completed a standardised 10 min warm  
145 up consisting of 4 min running at  $8\text{km}\cdot\text{h}^{-1}$ , 3 min of dynamic stretches (mainly focusing on the  
146 lower body) and a further 2 min of sub-maximal running before a final minute for the  
147 participant to perform their own stretches. Following this, and a full description of the test and  
148 safety procedures, participants began to run at a speed of  $8\text{km}\cdot\text{h}^{-1}$  at a 1% incline (21) on a  
149 motorised treadmill (HP Cosmos, Pulsar, Sportgerate GmbH, Nussdorf, Germany). The speed

150 of the treadmill was increased by  $1\text{km}\cdot\text{h}^{-1}$  every two minutes, this continued until participants  
151 reached 90% of their age predicted heart rate max ( $207 - (0.7 \times \text{age})$ ) (16). At this point, the  
152 treadmill speed remained constant whilst the incline of the treadmill was increased by 1% every  
153 minute until volitional exhaustion; this procedure was employed to avoid over-striding and  
154 potential early termination and inaccurate assessment of participant's  $\text{VO}_{2\text{peak}}$ . This method has  
155 been successfully used before to elicit  $\text{VO}_{2\text{peak}}$  during treadmill running in young athletes (40).  
156 Rolling 15 sec averages were calculated for the final minute of the test, with the highest 15 sec  
157 average during the test being regarded as  $\text{VO}_{2\text{peak}}$  (5).

158

159 The gaseous exchange threshold (GET) was identified using the V-slope method ( $V_E$  (ordinate),  
160  $\text{VO}_2$  (abscissa). Two regression lines were created based upon the relationship between  $\text{VCO}_2$   
161 and  $\text{VO}_2$ . The intercept point between the two regression lines was then visually identified,  
162 with the  $\text{VO}_2$  value at the intercept (GET) being extrapolated to the abscissa. To identify the  
163 speed at GET, a regression line was formulated for  $\text{VO}_2$  and running velocity, for each  
164 individual. The GET was assessed by two individual researchers (experienced in the detection  
165 of GET), demonstrating 81% agreement (17 out of 21). For the remaining four participants, a  
166 third researcher, also experienced in the detection of GET, was approached to verify the GET  
167 (15). The individual's  $\text{VO}_2$  at GET was then inputted into the individual's respective regression  
168 equation to calculate the running velocity at GET. The V-slope method has been shown to be  
169 a reliable method for detecting and identifying the gaseous exchange threshold in children (15).

170

### 171 *Assessment of Running Economy*

172 On the subsequent visit to the laboratory, and following a minimum of 24 hours recovery,  
173 participants' RE was assessed at three progressive sub-maximal intensities (one absolute:



174 8km·h<sup>-1</sup> and two relative: 80%GET and 95%GET exercise intensities), with 2 min passive  
175 recovery between each intensity. Prior to testing, participants were fitted with a Polar Heart  
176 rate monitor and face-mask, which was connected to the pre-calibrated online gas analysis  
177 (Cortex) system. Following a standardised 10 min warm-up and a full description of the test  
178 procedures, participants began to run at the lowest exercise intensity (for 3 of the participants  
179 the velocity at 80%GET was lower than 8km·h<sup>-1</sup>), with the gradient of the treadmill set at 1%  
180 (21). Participants ran at this speed for 4 min, ensuring that a steady state was maintained for  
181 the final minute (8). A 2 min passive recovery was then permitted, in which participants' heart  
182 rate recovered to within 10% of pre-exercise levels. Pilot testing demonstrated that a 2 min  
183 passive recovery was shown to provide sufficient time for players to recover, ensuring there  
184 were no ensuing effects on the subsequent exercise bouts. During this recovery period, the  
185 treadmill speed was increased to the next progressive speed (higher intensity of 8km·h<sup>-1</sup> or  
186 80%GET). Again, participants exercised at this speed for 4 min, attaining a steady state, before  
187 undertaking a further 2 min recovery. Following the final 2 min of recovery, participants  
188 completed another 4 min period of sub-maximal running at the highest intensity (95%GET).  
189 Extraction of the appropriate measures for assessing RE included applying rolling 15 sec  
190 averages for the final minute of each sub-maximal exercise intensity to ensure that a steady  
191 state had been attained. Current results revealed that all participants (100%) attained a steady  
192 state (< 2 mL·kg<sup>-1</sup>·min<sup>-1</sup>, in the final minute of exercise) for each sub-maximal exercise  
193 intensity. Following the identification of a steady state, RE was defined as the average VO<sub>2</sub>  
194 during the final minute of each exercise bout (8). Relative oxygen consumption (VO<sub>2</sub>) was  
195 obtained for each exercise intensity as well as determinants of RE: minute ventilation (V<sub>E</sub>) and  
196 ventilatory equivalent (V<sub>E</sub>VO<sub>2</sub>).

197

198 Measures of  $VO_2$  in both the maximal graded exercise test and the assessment of running  
199 economy, for each sub-maximal running speeds, were normalized using 1) ratio-standard  
200 scaling to body mass ( $\text{mL}\cdot\text{kg BM}^{-1}\cdot\text{min}^{-1}$ ) and fat free mass ( $\text{mL}\cdot\text{kg FFM}^{-1}\cdot\text{min}^{-1}$ ), 2) allometric  
201 scaling, using theoretically derived exponents (9, 25;  $b = 0.67$  and  $b = 0.75$ ) and, 3) a measure  
202 relative to participants'  $VO_{2\text{peak}}$  ( $\%VO_{2\text{peak}}$ ).

203

#### 204 *Assessment of oxygen uptake kinetics*

205 On the two remaining visits to the laboratory, each participant completed a series of exercise  
206 transitions to a higher intensity on the motorised treadmill. Each series consisted of 3 min  
207 unloaded (standing), 4 min running at an intensity equivalent to 95% GET (moderate intensity)  
208 and a run to exhaustion at an intensity equivalent to 60% $\Delta$  (severe intensity). Prior to the test,  
209 participants were familiarised with the transition to each speed given sufficient time to practise  
210 until they felt comfortable with both transitions (unloaded – moderate and moderate – severe).  
211 Following familiarisation to the speed transitions, participants were fitted with a Polar Heart  
212 rate monitor and face mask, which was connected to an online expired gas analysis system.  
213 During the test, verbal encouragement was provided throughout for participants to continue for  
214 as long as possible, however no visual feedback relating to exercise duration, was given to the  
215 participants during the test.

216

217 For the unloaded to moderate transition the treadmill was set at the relevant intensity for each  
218 individual, while the participant straddled the treadmill. The participant was then given a 10  
219 second countdown at the end of the unloaded phase, at which point they lowered themselves  
220 onto the moving treadmill and began exercising. For the transition from moderate to severe  
221 intensity the participant remained running on the treadmill. The time taken for each exercise  
222 transition was in all cases  $< 5$  seconds, thus having minimal effects on the  $VO_2$  kinetic response

223 as this would be contained within the cardiodynamic phase of the oxygen uptake response to  
224 an increase in intensity (40).

225

### 226 *Mathematical modelling of oxygen uptake kinetics*

227 Prior to the modelling of the  $\text{VO}_2$  kinetics errant breaths (coughing, swallowing, sighing, etc.)  
228 were removed from the raw data set so as not to distort or skew the underlying physiological  
229 response. Errant breaths were defined as a breath that was different to the mean of the  
230 surrounding four breaths by more than four times the standard deviation of the same  
231 surrounding four data points (22). Both data-sets from each stage (unloaded – moderate and  
232 moderate – severe) were time aligned and ensemble averaged to enhance the underlying  
233 physiological response characteristics for all intensities. Each ensemble average was then  
234 linearly interpolated second-by-second prior to the modelling process. Custom written software  
235 in Microsoft Excel, using the Solver function, was utilised for all modelling processes.

236

237 Pulmonary oxygen uptake kinetics for the unloaded to moderate and moderate to severe  
238 transitions were modelled separately, due to the difference in the characteristics of the kinetic  
239 response for each increase in intensity. For both, unloaded – moderate and moderate – to severe  
240 transitions, the goal of the modelling process was to isolate the fundamental phase (39) of  $\text{VO}_2$   
241 kinetic response (equation 1). To eliminate the influence of the cardiodynamic phase on the  
242 modelling of  $\text{VO}_2$  kinetics, the initial 20 seconds from the unloaded to moderate phase and  
243 initial 15 seconds of the moderate to severe phase were removed prior to the modelling process.  
244 A time delay of 20 seconds is often employed to accommodate the cardiodynamic phase (39);  
245 however, a 15 second time delay was adopted for the moderate to severe transition due to  
246 elevated baseline blood flow incurred from the prior moderate intensity (7). Following the

247 cardiodynamic phase,  $VO_2$  kinetics were assumed to develop initially via a single exponential  
248 term (fundamental phase), following a delay relative to the start of exercise of the form:

249

$$250 \quad V_{O_{2(t)}} = V_{O_{2(b)}} + A_{VO_2} \times (1 - \exp^{-(t-TD)/\tau}) \quad [1]$$

251

252 Where  $V_{O_{2(b)}}$  is the baseline  $VO_2$ , which was taken as the last 30 seconds of oxygen uptake  
253 during 3 minutes unloaded phase.  $A_{VO_2}$  represents the asymptotic amplitude of the fundamental  
254 component of the response;  $\tau$  is the time constant of the fundamental component and  $TD$  is the  
255 time delay similar, but not equal to the cardiodynamic-fundamental phase transition time. For  
256 the unloaded – moderate transition, the fundamental phase was considered *a priori* to  
257 encapsulate the entire 4-minute transition since exercise was undertaken below the GET. For  
258 the moderate – severe transition, the fitting strategy was designed to identify the onset of the  
259 “slow component” of the response to exercise, and thus isolate the fundamental component.  
260 Starting at 60 s, the fitting window was therefore widened by 1 s until the end of exercise with  
261 the time constant and reduced chi-square value of the curve of best fit for each time window  
262 plotted against time. The onset of the slow component could then be identified as the coincident  
263 point at which a plateau or minima in the value of  $\tau$  and a minima in chi-square, followed by a  
264 progressive increase in these values, could be determined as its value becomes affected by the  
265 slow component. The time at which this occurred was used as the optimal fitting window with  
266 which to determine the kinetics of the fundamental phase of  $VO_2$  kinetics. The phase III of  
267 oxygen uptake (steady state in the unloaded – moderate transition) was taken as the sum of  
268  $V_{O_{2(b)}}$  and  $A_{VO_2}$ . The amplitude of the slow component during the moderate – severe transition  
269 was calculated as  $VO_2$  at exhaustion minus the phase III  $VO_2$ .

270

271 *Yo-Yo Intermittent Recovery Test Level 1*

272 For the Yo-Yo IR1 test, cones were placed 20 m apart, with a 5 m recovery zone marked out  
273 at one end. The Yo-Yo IR1 test requires participants to run 2 x 20 m shuttle runs at increasing  
274 speeds, interspersed with 10 seconds of active recovery. The pace of the test was controlled by  
275 audio signals emitted from a CD player (Sony CFD-V7, Sony, Tokyo, Japan). For the maximal  
276 Yo-Yo IR1 test players were required to run until volitional termination of the test or, when  
277 they have twice failed to meet the designated cones in time with the audio signal, at which  
278 point they are removed from the test. The test score is the distance covered at the point they  
279 withdraw from the test. During the test, players were allowed to consume fluids ad libitum.  
280 Current findings support the use of the Yo-Yo IR1 test as a valid measure of physical  
281 performance, associated with soccer match-play, particularly within youth populations (6).

282

283 *Statistical Analysis*

284 A Shapiro Wilks test found the data to be normally distributed, therefore parametric statistical  
285 calculations were applied. Following this, independent *t*-tests were conducted to assess the  
286 differences between pre-PHV and mid-PHV groups, in anthropometric measures and measures  
287 of cardio-respiratory fitness. Differences between groups were also compared using Cohen's *d*  
288 effect sizes (ES) and thresholds (<0.5 = small; 0.5-0.8 = moderate; >0.8 = large) to aid  
289 interpretation. Additionally, where appropriate, a qualitative descriptor, used to aid practical  
290 inferences, was assigned to the following quantitative chances of benefit: 25-75% = benefit  
291 possible; 75-95% = benefit likely; 95-99% = benefit most likely; >99% = benefit almost  
292 certain. All statistical analysis was performed using SPSS version 21.0 (IBM SPSS statistics  
293 for Windows, IBM, Armonk, New York) and Microsoft Excel (Microsoft Excel 2013,  
294 Microsoft, Redmond, Washington) with the level of significance (alpha) set at 0.05.

295

296 **Results**

297 Differences in anthropometric and descriptive measurements revealed significant differences  
298 and large effect sizes for stature, body mass, fat free mass and maturity offset, with the mid-  
299 PHV group presenting greater values than the pre-PHV group, despite being of similar  
300 chronological age (Table 2). Furthermore, years spent training also revealed significant and  
301 large effect sizes between groups, but with the pre-PHV group demonstrating greater values  
302 than the mid-PHV group, meaning that those identified as pre-PHV had reported to have spent  
303 a greater amount of years of systematic training at a high level than the mid-PHV group.

304

305 **Table 2:** Differences in anthropometric and descriptive characteristics between those identified  
306 as pre- and mid-PHV, in highly trained youth soccer players.

307 \*\*\* Insert Table 2 Here\*\*\*

308

309 While absolute measures ( $L \cdot \text{min}^{-1}$ ) of  $VO_{2\text{ peak}}$  were shown to be greater in the mid-PHV group,  
310 the difference between the two groups was eradicated when expressed relative to body mass,  
311 fat free mass, as a  $\%VO_{2\text{ peak}}$  or when using theoretically derived exponents (Table 3). In  
312 addition, a moderate effect size was revealed for the velocity at GET, with the mid-PHV group  
313 demonstrating a faster velocity when running at GET.

314

315 Limited differences were evident between the two groups for both, measures of cardio-  
316 respiratory fitness and determinants of running economy when running at an absolute speed of  
317  $8\text{km} \cdot \text{h}^{-1}$ . Similarly, no differences in running economy were evident for each submaximal  
318 running speed when  $VO_2$  was expressed in relation to body mass or fat free mass. For both the  
319 relative submaximal exercise intensities, however, the pre-PHV group presented significantly  
320 superior levels of running economy, when  $VO_2$  data was scaled using theoretically derived

321 exponents. In support of this, the pre-PHV group also demonstrated a reduced fractional  
322 utilisation ( $\%VO_{2\text{peak}}$ ) for the same relative submaximal exercise intensities (Table 3). Figure  
323 1 displays a representative plot of the  $VO_2$  for each of the three sub-maximal running speeds  
324 and during each of the 2 min passive recovery phases.

325

326 **Table 3:** Differences between those identified as pre-PHV and mid-PHV, in measures of  
327 cardio-respiratory fitness during the maximal graded exercise test and during both absolute  
328 ( $8\text{km}\cdot\text{h}^{-1}$ ) and relative (80% and 95% GET) submaximal running speeds, using different  
329 methods for normalizing  $VO_2$ .

330 \*\*\*Insert Table 3 Here\*\*\*

331

332 **Figure 1:** Representative plot of the  $VO_2$  for each of the three sub-maximal running speeds and  
333 during each of the 2 min passive recovery phases

334 \*\*\*Insert Fig 1 Here\*\*\*

335

336 Measures of  $VO_2$  kinetics revealed a significant difference for the absolute amplitude (L.min)  
337 and a large effect size for the phase II  $\tau$ , during the low – mod transition, with the pre-PHV  
338 group demonstrating faster oxygen uptake kinetics than the mid-PHV group. Figure 2 displays  
339 a representative plot of the pulmonary oxygen uptake kinetics during the work-to-work  
340 protocol.

341

342 **Figure 2:** Representative plot of the pulmonary oxygen uptake kinetics during the work-to-  
343 work protocol, with the respective unloaded - moderate and moderate - severe time constants  
344 ( $\tau$ s) (Doncaster et al., 2016).

345

\*\*\*Insert Fig 2 Here\*\*\*

346 Finally, performance in the Yo-Yo IR1, a measure of soccer-specific endurance, was similar  
347 between the two groups (mid-PHV:  $1681 \pm 304$  vs. pre-PHV:  $1572 \pm 360$  m,  $P = 0.49$ ,  $d =$   
348  $0.33$ ), revealing a small effect size.

349

## 350 **Discussion**

351 The main finding from the current study was that highly trained soccer players of advanced  
352 maturity status do not outperform their less mature counterparts, in key measures of cardio-  
353 respiratory fitness and determinants of running economy. Instead, results show that players  
354 identified as less mature (pre-PHV) display enhanced levels of cardio-respiratory fitness,  
355 particularly when accounting for differences in body size.

356

357 In line with the existing literature (24, 26, 27), anthropometric and descriptive characteristics  
358 found those players of an advanced maturity status (mid-PHV) to be physically taller and  
359 bigger, presenting both a larger stature, body mass and estimated fat free mass than their less  
360 mature counterparts (pre-PHV) of a similar age. The pre-PHV group, however, reported a  
361 greater amount of training experience at a high level than their more mature counterparts.  
362 Despite those identified as mid-PHV displaying a significantly greater absolute measure of  
363  $VO_{2peak}$ , when these data were expressed relative to: body mass ( $mL \cdot kg \text{ BM}^{-1} \cdot \text{min}^{-1}$ ), fat free  
364 mass ( $mL \cdot kg \text{ FFM}^{-1} \cdot \text{min}^{-1}$ ) or when using theoretical exponents ( $mL \cdot kg^{-0.67} \cdot \text{min}^{-1}$  and  $mL \cdot kg^{-$   
365  $0.75} \cdot \text{min}^{-1}$ ), the differences between the two groups were eliminated (Table 3). Similarly,  
366 performance during the Yo-Yo IR1 test was comparable between the two groups, revealing  
367 only a small effect size in the favour of the mid-PHV group. Results, however, do propose a  
368 preference for oxidative metabolism in less mature players (pre-PHV), when compared to their  
369 more mature counterparts (mid-PHV). Indeed, when running at relative sub-maximal  
370 intensities (80% & 95% GET), normalized  $VO_2$  values using theoretically derived exponents



371 ( $b = 0.67$  and  $b = 0.75$ ), the pre-PHV group revealed a significantly reduced  $VO_2$  response for  
372 a given relative sub-maximal intensity. In support of this, the pre-PHV group also presented a  
373 reduced fractional utilisation ( $\%VO_{2peak}$ ) when running at relative sub-maximal intensities  
374 (80% & 95% GET), as well as faster  $VO_2$  kinetics during low – moderate exercise transitions  
375 ( $\tau$ ), in comparison to their more mature counterparts.

376

377 The results obtained from the tests employed within the current study would appear to be  
378 indicative of highly trained youth soccer players. Relative measures of  $VO_{2peak}$  in the current  
379 study are in-line with those of Cunha et al. (9), who reported values of  $59.6 \pm 4.3 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$   
380 <sup>1</sup> in pubescent (Age:  $13.4 \pm 1.0$  yr, Body Mass:  $62.5 \pm 9.9$  kg) soccer players and Le Gall et al.  
381 (24), who reported values of  $59.2 \pm 3.2 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$  in U14 international players (Age:  $13.4$   
382  $\pm 0.4$  yr, Body Mass:  $52.5 \pm 9.9$  kg). Similarly, players' performance in the Yo-Yo IR1 test  
383 within the current study is greater than figures reported within the literature for players of a  
384 similar age and standard of performance. Indeed, Deprez et al. (11) and Deprez et al. (13)  
385 reported scores of  $1319 \pm 366$  m and  $1270 \pm 440$  m for U13 high level youth soccer players,  
386 respectively. Deprez et al. (11), however, did separate their cohort of U13 players into low,  
387 average and high performers and reported scores of  $886 \pm 114$  m,  $1357 \pm 100$  m and  $1714 \pm$   
388  $145$  m in the Yo-Yo IR1, respectively. In this regard players' performance in the current study  
389 of  $1682 \pm 304$  m for the mid-PHV group and  $1572 \pm 360$  m for pre-PHV group, could be  
390 regarded as above average (11). Nevertheless, the current data demonstrates that the players  
391 recruited within this study are indicative and therefore representative, of highly trained youth  
392 soccer players.

393

394 Current results are in accordance with Cunha et al. (9) who also found that while those of  
395 advanced maturity present higher absolute values ( $\text{L}\cdot\text{min}^{-1}$ ) of  $VO_{2max}$ , when expressed relative

396 to body mass ( $\text{mL}\cdot\text{kg BM}^{-1}\cdot\text{min}^{-1}$ ) differences in measures of  $\text{VO}_{2\text{peak}}$  are removed (10, 25). As  
397 in the present study, Cunha et al. (9) also analysed their data using theoretical exponents ( $b =$   
398  $0.67$  and  $b = 0.75$ ), as well as their own experimentally derived exponent ( $b = 0.90$ ), using  
399 linear regression analysis, to scale participants'  $\text{VO}_2$  response. While Cunha et al. (9) reported  
400 a significant difference between pre- and post-pubescent youth soccer players, they also  
401 reported that differences between groups could not be attributed to biological maturation in the  
402 multiple linear regression analysis. Initial findings (2, 3, 4), examining the impact of maturation  
403 on  $\text{VO}_{2\text{max}}$  reported that maturation positively affected measures of  $\text{VO}_{2\text{max}}$ , however, authors  
404 only expressed measures of  $\text{VO}_{2\text{max}}$  in absolute terms. In line with the present findings, recent  
405 research (9, 10, 25) advocates the need for appropriate analytical procedures (scaling  
406 techniques using lower-limb muscle volume) when assessing differences in  $\text{VO}_2$  within  
407 heterogeneous populations with respect to maturity status. Appropriately interpreted measures  
408 of  $\text{VO}_{2\text{peak}}$  are unaffected by maturation in highly trained youth soccer players. Whether or not  
409 this has implications for physical performance during soccer match-play requires further  
410 investigation.

411

412 Present results suggest a reduced oxygen cost for a given relative submaximal running  
413 intensity, in players identified as pre-PHV, when compared to their more mature counterparts.  
414 There is a paucity of research to date which has examined differences in running economy  
415 between individuals of different maturity status, particularly in team sport athletes. Segers et  
416 al. (34), reported no differences in the oxygen cost (expressed in absolute terms, using  
417 allometric scaling ( $b = 0.59$ ) or as a  $\% \text{VO}_{2\text{peak}}$ ) of running between youth soccer players who  
418 were identified as either 'early' or 'late' maturers, when running at absolute speeds of 8, 9.5  
419 and  $11\text{km}\cdot\text{h}^{-1}$  on a treadmill. The use of absolute running speeds however, in the study of Segers  
420 et al. (34) may be inappropriate for youth populations as it is likely to result in different internal

421 responses, leading to large inter-individual variability and subsequently increased standard  
422 deviations, masking any inter-group differences (32). The use of relative running intensities  
423 however, will reduce the inter-individual variability and is a more appropriate method by which  
424 youth soccer players' running economy can be assessed, in relation to maturity status. Indeed,  
425 the improved levels of running economy and fractional utilisation at relative submaximal  
426 intensities, in the present study, elucidates to an improved ability to utilise the aerobic  
427 metabolism in those identified as pre-PHV, potentially compensating for the undeveloped  
428 anaerobic metabolism (24, 27, 33), within the pre-PHV group. Ratel (33) suggests that younger  
429 populations (e.g. children) have a reduced capacity for anaerobic activity (lower concentrations  
430 of glycolytic enzyme activity [phosphofructokinase], lower muscle mass, reduced percentage  
431 of type II muscle fibres) but an improved capacity for aerobic energy production (increased  
432 oxidative phosphorylation enzyme activity [citrate synthase], increased percentage of type I  
433 muscle fibres). The extent to which these physiological mechanisms are affected by high levels  
434 of systematic team sport training requires further investigation.

435

436 Another means of evaluating an individual's aerobic fitness is via the assessment of  $VO_2$   
437 kinetics. The relative contribution of oxidative and non-oxidative energy supply during  
438 exercise are dependent upon the characteristics of the  $VO_2$  response to exercise, and is regarded  
439 as an important indicator of aerobic function that influences an individual's capacity to perform  
440 (22). Current results provide evidence to suggest superior  $VO_2$  kinetics (faster *taus*) during a  
441 low to moderate exercise transition, but not during a moderate to severe transition, in youth  
442 soccer players identified as pre-PHV. Indeed, faster  $VO_2$  kinetics (*tau*) during low to moderate  
443 exercise transitions have been shown to be correlated with the volume and maintenance of high  
444 speed activities during youth soccer match-play (14). With faster *taus* relating to a greater  
445 volume and an improved ability to maintain high speed activities during youth soccer match-

446 play. Furthermore, research into adolescent male and female soccer players has demonstrated  
447 faster  $VO_2$  kinetics in soccer trained individuals in comparison to their untrained counterparts  
448 (29, 37). Current results also support previous research which highlights faster *taus* in younger  
449 populations, with such measures becoming progressively slower (i.e. larger *taus*) as individuals  
450 transition from childhood, through adolescence and then to adulthood (23). Nevertheless, it  
451 should be acknowledged that players'  $VO_2$  kinetics profiles were averaged from two transitions  
452 and that, where possible, multiple transitions should be encouraged as this will improve  
453 confidence in the model parameter estimates (e.g. *tau*) (22).

454

455 In contrast to measures of anaerobic fitness components, such as speed, power and strength  
456 (26, 27), where those of an advanced maturity status are shown to excel, measures of aerobic  
457 fitness are shown to be either, similar between groups of differing maturity status or favour less  
458 mature individuals, when appropriately scaled (9, 10, 25). This is supported by the present  
459 result, which show no differences between pre- and mid-PHV groups in measures of soccer-  
460 specific endurance (Yo-Yo IR1), as well as the absence of any relationship between players'  
461 maturity offset and Yo-Yo IR1 performance ( $r = 0.16$ ,  $P > 0.05$ ). As noted by Malina et al. (27)  
462 the years spent in formal training was a significant contributor to intermittent endurance  
463 capacity (measured via the Yo-Yo IR1) in a cohort of 13-15 yr old, high level youth soccer  
464 players. Therefore, as those in the pre-PHV group reported to have spent significantly more  
465 years training than their more mature counterparts (pre-PHV:  $5.9 \pm 1.8$  vs. mid-PHV:  $3.7 \pm 2.0$   
466 yr,  $P < 0.05$ ,  $d = 1.16$ ), any maturational differences between the two groups, in the current  
467 results, may have been offset by the greater level of experienced training within the pre-PHV  
468 group. Furthermore, Lovell et al. (26) reported trivial and small effect sizes in the Multi-stage  
469 fitness test (MSFT), between players born in the first quarter of the year (Sept-Nov) and players  
470 born in the final quarter of the year (June-Aug) in U13-U16 English academy youth soccer

471 players. Although, chronological age rather than maturity status was used to separate groups in  
472 the study of Lovell et al. (26).

473

474 Despite the consensus for appropriate allometric scaling within the literature, and in particular  
475 the support for experimentally derived components (9, 25), the current study only employed a  
476 small sample size. Indeed, research suggests that relatively large sample sizes are required to  
477 lower statistical inference errors, and that factors such as ‘quality’ and ‘quantity’ of the data,  
478 as well as analysis method (e.g. nonlinear regression or linear regressions) are likely to  
479 influence the allometric scaling exponent beta estimation (18). Given the small samples (pre-  
480 PHV:  $n = 10$ , mid-PHV:  $n = 11$ ) within the current study, an experimentally observed exponent,  
481 for allometric scaling, was not possible. As a result, theoretical exponents ( $b = 0.67$  and  $b =$   
482  $0.75$ ), estimates of Fat Free Mass and a fractional utilisation measure ( $\%VO_{2peak}$ ) were  
483 employed to assess highly trained youth soccer players’  $VO_2$  responses, in relation to maturity  
484 status (i.e. pre-PHV vs. mid-PHV). Future research, however, where possible, should look to  
485 develop their own experimentally observed exponents, for allometric scaling. Furthermore,  
486 limitations associated with the calculations used for estimating body fat percentage, and  
487 therefore FFM should also be acknowledged. Mainly the appropriateness of the algorithms  
488 used (35), to the elite youth soccer players should be considered. Nevertheless, in the absence  
489 of population specific algorithms, previous research examining body composition in  
490 professional soccer players have employed these methods (20). Despite these limitations the  
491 current study extends and improves upon the existing research by examining differences in  
492 distinct measures of cardio-respiratory fitness, and determinants of running economy with  
493 respect to maturity status and in a group of highly trained youth soccer players, in which there  
494 has been an attempt to account for the effects of body size on  $VO_2$ .

495

496 While the present results do advocate the need for an enhanced aerobic capacity in highly  
497 trained youth soccer players that are identified as pre-PHV. They also demonstrate that, youth  
498 academy soccer players demonstrate high levels of soccer-specific endurance (Yo-Yo IR1),  
499 irrespective of maturity status (41). An enhanced capacity for aerobic metabolism, in those  
500 identified as pre-PHV may potentially transcend into levels of physical performance during  
501 high intensity intermittent exercise that are in comparison to their more mature counterparts.  
502 Nevertheless, coaches should be aware that potential differences in physical performance  
503 between players identified as pre-PHV or post-PHV are likely to be a consequence of anaerobic  
504 fitness capabilities and not aerobic fitness capabilities (24, 26, 41). An emphasis on such  
505 anaerobic fitness capabilities (e.g. strength and speed), may be one reason why there is a  
506 selection bias toward players of an advanced maturity status within these age categories (12-  
507 14 yrs old), often referred to as the relative age effect within soccer (12, 17, 26).

508

## 509 **Conclusion**

510 In summary, current findings provide further evidence for differences in anthropometric and  
511 descriptive measures between players identified as pre-PHV and players identified as post-  
512 PHV, despite similar chronological age. The present data, however, also demonstrates that  
513 players of an advanced maturity status (mid-PHV) do not outperform their less mature (pre-  
514 PHV) counterparts in a range of measures and determinants of aerobic fitness. Rather, the  
515 current data would suggest that those identified as pre-PHV have a superior ability to utilise  
516 aerobic energy pathways during relative submaximal intensities, while also displaying faster  
517  $VO_2$  kinetics (*tau*) when transitioning from a low to moderate exercise intensity. Despite the  
518 potential for improved capacity for oxidative metabolism in the pre-PHV group, soccer-  
519 specific endurance performance (Yo-Yo IR1) was similar between the two groups.

520 Nevertheless, these enhanced levels of cardio-respiratory fitness may partly explain the  
521 comparable levels of high intensity-intermittent performance during the Yo-Yo IR1 test.

522

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527

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643 **Table 1:** Anthropometric and screening measures ( $n = 21$ )

Variable	Mean $\pm$ Standard Deviation	90% Confidence Limits
Age (y)	13.2 $\pm$ 0.6	12.9 - 13.4
Stature (m)	1.59 $\pm$ 0.09	1.54 - 1.62
Body Mass (kg)	48.0 $\pm$ 10.2	43.9 - 52.4
Maturity Offset (y)	-0.8 $\pm$ 0.9	-1.2 to -0.4
Tanner Stage	3 $\pm$ 1	2 - 3
Predicted Age at PHV	14.0 $\pm$ 0.7	13.7 - 14.3
$\Sigma$ 4 Skinfolds (mm)	30.2 $\pm$ 5.4	28.0 - 32.4
Body Fat (%)	17.6 $\pm$ 2.2	16.7 - 18.6
Fat Free Mass (kg)	39.6 $\pm$ 8.7	35.8 - 43.3
Training Years (y)	4.8 $\pm$ 2.2	3.9 - 5.7
Training Hours (hrs.p.week)	12.4 $\pm$ 2.7	11.4 - 13.7

644 Note: PHV = Peak Height Velocity; Skinfold sites used for the  $\Sigma 4$  skinfolds were the biceps, triceps, subscapular  
 645 and superiliac.  
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658 **Table 2:** Table 2: Differences in anthropometric and descriptive characteristics between those identified as pre- and mid-PHV, in highly trained  
 659 youth soccer players.

	Mid-PHV ( <i>n</i> = 11)		Pre-PHV ( <i>n</i> = 10)		Difference		
	Mean ± SD	90% CL	Mean ± SD	90% CL	Sig.		Cohen's <i>d</i>
Age (yr)	13.4 ± 0.4	13.1 - 13.6	13.0 ± 0.7	12.5 - 13.4	<i>P</i> = 0.12	0.52	Moderate
Stature (m)	1.65 ± 0.08	1.60 - 1.69	1.52 ± 0.06	1.48 - 1.56	<i>P</i> < 0.05	1.83	Large
Body Mass (kg)	53.9 ± 10.7	48.2 - 60.5	41.6 ± 3.9	39.3 - 43.8	<i>P</i> < 0.05	1.53	Large
Tanner	3.2 ± 1.0	2.6 - 3.8	2.6 ± 0.5	2.3 - 2.9	<i>P</i> = 0.11	0.77	Moderate
Maturity Offset	-0.2 ± 0.7	-0.6 to 0.2	-1.5 ± 0.4	-1.7 to -1.3	<i>P</i> < 0.05	2.28	Large
Predicted Age at PHV	13.6 ± 0.6	13.2 - 13.9	14.4 ± 0.5	14.1 - 14.7	<i>P</i> < 0.05	1.45	Large
Σ Skinfolds (mm)	28.9 ± 4.2	26.6 - 31.4	31.6 ± 6.5	27.6 - 35.4	<i>P</i> = 0.26	0.49	Small
Body Fat (%)	17.2 ± 1.7	16.2 - 18.2	18.1 ± 2.6	16.5 - 19.8	<i>P</i> = 0.32	0.47	Small
Fat Free Mass (kg)	44.7 ± 9.3	39.2 - 50.2	34.0 ± 2.7	32.3 - 35.6	<i>P</i> < 0.05	1.62	Large
Training Years	3.7 ± 2.0	2.5 - 4.9	5.9 ± 1.8	4.8 - 6.9	<i>P</i> < 0.05	1.16	Large
Training Hours (per week)	12.3 ± 1.4	11.5 - 13.1	12.5 ± 3.7	10.3 - 15.1	<i>P</i> = 0.85	0.07	Trivial

660 Note: PHV = Peak Height Velocity; Skinfold sites used for the Σ4 skinfolds were the biceps, triceps, subscapular and superiliac.

661

662 **Table 3:** Differences between those identified as pre-PHV and mid-PHV, in measures of cardio-respiratory fitness during the maximal graded  
 663 exercise and during both absolute (8km·h<sup>-1</sup>) and relative (80% and 95% GET) submaximal running speeds, using different methods for normalizing  
 664 VO<sub>2</sub>.  
 665

	Mid-PHV ( <i>n</i> = 11)		Pre-PHV ( <i>n</i> = 10)		90% CI of Differences			Between Group Differences		Qualitative Descriptor
	Mean ± SD	90% CL	Mean ± SD	90% CL	Lower	Upper	Sig.	Cohen's <i>d</i>		
VO <sub>2peak</sub> (L·min <sup>-1</sup> )	3.09 ± 0.53	2.80 - 3.42	2.50 ± 0.29	2.31 - 2.67	-0.96	-0.25	<i>P</i> = 0.005	1.38	Large	Very Likely
VO <sub>2max</sub> (mL·kg BM <sup>-1</sup> ·min <sup>-1</sup> )	58.6 ± 5.7	55.3 - 61.6	60.2 ± 4.9	57.3 - 63.2	-2.4	6.1	<i>P</i> = 0.49	0.30	Small	Unclear
VO <sub>2peak</sub> (mL·kg FFM <sup>-1</sup> ·min <sup>-1</sup> )	70.1 ± 7.3	65.8 - 74.3	73.5 ± 5.4	70.1 - 76.9	-1.4	8.3	<i>P</i> = 0.24	0.56	Moderate	Unclear
VO <sub>2peak</sub> (mL·kg <sup>-0.67</sup> ·min <sup>-1</sup> )	214.9 ± 21.0	202.5 - 227.3	205.6 ± 17.3	194.9 - 216.3	-23.9	5.3	<i>P</i> = 0.29	0.50	Moderate	Unclear
VO <sub>2peak</sub> (mL·kg <sup>-0.75</sup> ·min <sup>-1</sup> )	156.4 ± 15.1	147.5 - 165.4	152.6 ± 12.5	144.9 - 160.4	-14.3	6.7	<i>P</i> = 0.54	0.28	Small	Unclear
Max V <sub>E</sub> VO <sub>2</sub>	32.0 ± 4.1	29.8 - 34.7	30.7 ± 1.7	29.7 - 31.8	-4.2	1.1	<i>P</i> = 0.36	0.41	Small	Unclear
Velocity at GET (km·h <sup>-1</sup> )	11.4 ± 1.0	10.8 - 11.9	10.7 ± 0.8	10.2 - 11.2	-1.5	0.1	<i>P</i> = 0.10	0.77	Moderate	Likely
VO <sub>2</sub> at GET (% VO <sub>2peak</sub> )	81.9 ± 4.9	79.2 - 84.6	82.3 ± 4.2	79.8 - 85.1	-3.4	4.2	<i>P</i> = 0.83	0.08	Trivial	Unclear
<b>Running Economy</b>										
VO <sub>2</sub> @ 8km/h (mL·kg BM <sup>-1</sup> ·min <sup>-1</sup> )	39.6 ± 4.2	37.3 - 41.9	39.4 ± 4.6	36.7 - 42.4	-3.9	3.4	<i>P</i> = 0.94	0.05	Trivial	Unclear
VO <sub>2</sub> @ 8km/h (mL·kg FFM <sup>-1</sup> ·min <sup>-1</sup> )	47.8 ± 4.6	45.0 - 50.5	48.1 ± 5.4	44.8 - 51.5	-3.4	4.2	<i>P</i> = 0.87	0.08	Trivial	Unclear
VO <sub>2</sub> @ 8km/h (mL·kg <sup>-0.67</sup> ·min <sup>-1</sup> )	146.9 ± 16.3	137.3 - 156.5	134.7 ± 16.5	124.5 - 144.9	-24.5	0.2	<i>P</i> = 0.11	0.78	Moderate	Likely
VO <sub>2</sub> @ 8km/h (mL·kg <sup>-0.75</sup> ·min <sup>-1</sup> )	106.8 ± 11.3	100.1 - 113.6	100.0 ± 12.0	92.5 - 107.4	-15.7	1.9	<i>P</i> = 0.20	0.62	Moderate	Likely
V <sub>E</sub> VO <sub>2</sub> @ 8km·h <sup>-1</sup>	26.2 ± 2.4	24.8 - 27.6	27.6 ± 1.6	26.7 - 28.8	-0.4	3.2	<i>P</i> = 0.14	0.69	Moderate	Likely
RER @ 8km·h <sup>-1</sup>	0.89 ± 0.04	0.86 - 0.91	0.90 ± 0.03	0.88 - 0.92	-0.02	0.04	<i>P</i> = 0.52	0.28	Small	Unclear
%HRmax @ 8km·h <sup>-1</sup>	77.1 ± 4.9	74.4 - 79.7	77.9 ± 5.4	74.7 - 81.1	-3.3	5.3	<i>P</i> = 0.70	0.15	Trivial	Unclear
%VO <sub>2peak</sub> @ 8km·h <sup>-1</sup>	67.7 ± 4.5	65.0 - 70.3	65.6 ± 7.7	61.2 - 70.4	-7.4	3.3	<i>P</i> = 0.46	0.33	Small	Unclear
VO <sub>2</sub> @ 80%GET (mL·kg BM <sup>-1</sup> ·min <sup>-1</sup> )	43.7 ± 5.1	41.0 - 47.0	42.2 ± 3.8	39.7 - 44.3	-5.4	1.9	<i>P</i> = 0.43	0.33	Small	Unclear
VO <sub>2</sub> @ 80%GET (mL·kg FFM <sup>-1</sup> ·min <sup>-1</sup> )	52.8 ± 5.9	49.3 - 56.3	51.7 ± 4.6	48.8 - 54.6	-5.2	2.9	<i>P</i> = 0.64	0.22	Small	Unclear
VO <sub>2</sub> @ 80%GET (mL·kg <sup>-0.67</sup> ·min <sup>-1</sup> )	162.2 ± 19.2	150.9 - 173.6	144.5 ± 13.4	136.2 - 152.8	-30.3	-5.1	<i>P</i> = 0.025	1.12	Large	Very Likely
VO <sub>2</sub> @ 80%GET (mL·kg <sup>-0.75</sup> ·min <sup>-1</sup> )	118.0 ± 13.5	110.0 - 126.0	107.3 ± 9.8	101.2 - 113.4	-19.7	-1.7	<i>P</i> = 0.053	0.95	Large	Likely
V <sub>E</sub> VO <sub>2</sub> @ 80%GET	26.9 ± 2.1	25.6 - 28.1	27.9 ± 1.5	27.0 - 28.8	-0.5	2.5	<i>P</i> = 0.28	0.55	Moderate	Unclear
RER @ 80%GET	0.90 ± 0.03	0.88 - 0.92	0.91 ± 0.03	0.89 - 0.92	-0.01	0.03	<i>P</i> = 0.45	0.33	Small	Unclear
%HRmax @ 80%GET	82.4 ± 4.9	79.1 - 84.8	82.4 ± 4.1	79.8 - 84.7	-3.8	4.1	<i>P</i> = 0.99	0.00	no effect	Unclear
%VO <sub>2peak</sub> @ 80%GET	74.8 ± 5.6	71.4 - 78.0	70.2 ± 4.5	67.4 - 72.8	-8.7	-0.2	<i>P</i> = 0.049	0.67	Moderate	Likely
VO <sub>2</sub> @ 95%GET (mL·kg BM <sup>-1</sup> ·min <sup>-1</sup> )	50.6 ± 5.0	47.9 - 53.4	48.6 ± 4.2	46.0 - 51.0	-5.8	1.7	<i>P</i> = 0.33	0.43	Small	Unclear
VO <sub>2</sub> @ 95%GET (mL·kg FFM <sup>-1</sup> ·min <sup>-1</sup> )	61.3 ± 5.6	58.0 - 64.6	59.6 ± 5.3	56.3 - 62.9	-5.8	2.4	<i>P</i> = 0.49	0.32	Small	Unclear
VO <sub>2</sub> @ 95%GET (mL·kg <sup>-0.67</sup> ·min <sup>-1</sup> )	188.3 ± 19.7	176.7 - 199.9	166.6 ± 15.2	157.2 - 176.0	-35.1	-8.3	<i>P</i> = 0.011	1.29	Large	Very Likely
VO <sub>2</sub> @ 95%GET (mL·kg <sup>-0.75</sup> ·min <sup>-1</sup> )	137.0 ± 13.5	129.0 - 144.9	123.7 ± 11.1	116.8 - 130.6	-22.7	-3.9	<i>P</i> = 0.024	1.13	Large	Very Likely
V <sub>E</sub> VO <sub>2</sub> @ 95%GET	27.5 ± 2.6	25.9 - 28.9	27.8 ± 2.3	26.5 - 29.1	-1.6	2.3	<i>P</i> = 0.77	0.12	Trivial	Unclear
RER @ 95%GET	0.93 ± 0.04	0.91 - 0.95	0.93 ± 0.04	0.90 - 0.96	-0.03	0.03	<i>P</i> = 0.99	0.00	no effect	Unclear
%HRmax @ 95%GET	88.7 ± 4.3	86.0 - 91.0	89.2 ± 3.9	86.5 - 91.4	-2.9	4.0	<i>P</i> = 0.77	0.12	Trivial	Unclear
%VO <sub>2peak</sub> @ 95%GET	86.6 ± 5.3	82.9 - 89.6	81.1 ± 7.4	75.9 - 85.0	-11.4	-0.1	<i>P</i> = 0.06	0.85	Large	Likely
<b>VO<sub>2</sub> Kinetics</b>										
low - mod <i>tau</i> (s)	22.0 ± 8.1	18.2 - 27.4	16.4 ± 5.6	13.6 - 20.0	-11.7	-0.06	<i>P</i> = 0.08	0.80	Large	Likely
low - mod amplitude (L·min <sup>-1</sup> )	1.91 ± 0.43	1.68 - 2.15	1.52 ± 0.19	1.41 - 1.63	-0.66	-0.14	<i>P</i> = 0.016	1.17	Large	Very Likely
mod - sev <i>tau</i> (s)	78.8 ± 61.0	47.1 - 119.6	47.6 ± 23.7	34.6 - 64.7	-73.4	2.9	<i>P</i> = 0.15	0.67	Moderate	Likely
mod - sev amplitude (L·min <sup>-1</sup> )	0.56 ± 0.21	0.44 - 0.69	0.44 ± 0.24	0.31 - 0.59	-0.32	0.07	<i>P</i> = 0.22	0.53	Moderate	Likely
<b>Yo-Yo IRI (m)</b>	1682 ± 304	1512 - 1855	1572 ± 360	1364 - 1798	-402	188	<i>P</i> = 0.49	0.33	Small	Unclear

666 Note:  $VO_{2peak}$ = peak oxygen consumption; GET= gaseous exchange threshold;  $V_EVO_2$ = ventilatory equivalent; RER= respiratory exchange ratio;  $HR_{max}$ = maximal heart rate;  
667  $\tau$ = time constant; Yo-Yo IR1= Yo-Yo intermittent recovery test level 1.