Maximal Fat Oxidation is Related to Performance in an Ironman Triathlon

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ABSTRACT

The aim of the present study was to investigate the relationship between maximal fat oxidation rate (MFO) measured during a progressive exercise test on a cycle ergometer and ultra-endurance performance. 61 male ironman athletes (age: 35 ± 1 yrs. [23–47 yrs.], with a BMI of 23.6 ± 0.3 kg/m^2 [20.0–30.1 kg/m^2], a body fat percentage of $16.7 \pm 0.7\%$ [8.4–30.7%] and a VO$_2$peak of 58.7 ± 0.7 ml/min/kg [43.9–72.5 ml/min/kg] SEM [Range]) were tested in the laboratory between 25 and 4 days prior to the ultra-endurance event, 2016 Ironman Copenhagen. Simple bivariate analyses revealed significant negative correlations between race time and MFO ($r^2 = 0.12$, $p < 0.005$) and VO$_2$peak ($r^2 = 0.45$, $p < 0.0001$) and a positive correlation between race time and body fat percentage ($r^2 = 0.27$, $p < 0.0001$). MFO and VO$_2$peak were not correlated. When the significant variables from the bivariate regression analyses were entered into the multiple regression models, VO$_2$peak and MFO together explained 50% of the variation observed in race time among the 61 Ironman athletes (adj $R^2 = 0.50$, $p < 0.001$). These results suggests that maximal fat oxidation rate exert an independent influence on ultra-endurance performance (>9h). Furthermore, we demonstrate that 50% of the variation in Ironman triathlon race time can be explained by peak oxygen uptake and maximal fat oxidation.

Introduction

Triathlon is a multistage competition involving three consecutive disciplines: swimming, cycling and running over a variety of distances. The Ironman distance was introduced in 1978 on the Hawai'ian island of O'ahu at Waikiki beach, it consists of, 3.8 km open water swim, 180 km bike ride, and a 42.2 km run. This extreme endurance event has since spread worldwide attracting thousands of endurance-trained athletes [21]. Studies have investigated the anthropometric characteristics of ironman athletes [14, 18] and the physiological stress observed in these athletes after an ironman-distance triathlon event [20, 22]. However, to our surprise, we have not found studies that directly investigated the relationship between the metabolic characteristics and overall race time in the ironman-distance triathlon.

Ever since the hallmarkstudy by Costill and colleagues, where they manipulated plasma substrate availability and demonstrated that high plasma fatty acid concentrations resulted in a glycogen sparing effect with a concomitant high fat oxidation during moderate intensity exercise [6], practical applications of this has been tested to enhance exercise performance. It is well known that carbohydrate stores are limited and that exogenous uptake cannot match utilization rates during prolonged moderate to high intensity exercise and this inevitably leads to muscle and liver glycogen depletion and thus fatigue and decreased performance, known as "hitting the wall". A number of approaches, high fat feeding, overnight fasted training, training with low glycogen content and recently keto-adaptation have been applied to achieve enhanced performance, but for performance times below 2–3 h, there is no solid

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evidence supporting any other procedure than the traditionally recommended high carbohydrate diet in athletes[4, 17]. Moreover, when performance times are above 4 h there are very few good studies linking performance and metabolism. Interestingly, a case study at the 2006 ironman world championships applied the doubly labelled water technique in one participant and found a total energy expenditure of 8,926 kcal of which 3,132 kcal was derived from fat oxidation [7]. Based on this we speculated that in addition to a high maximal oxygen uptake also a high fat oxidation capacity could be of major importance in endurance events that in duration are well above the 2–4 h, where performance cannot be sustained on glycogen.

Our aim was to investigate if maximal fat oxidation capacity was linked to performance in participants in an ironman triathlon. We hypothezied that a high maximal fat oxidation capacity measured by an incremental exercise cycle test would translate into a performance benefit and thus a faster racetime in an ironman-distance triathlon.

Methods

General design

Data were collected between 25 and four days before the Ironman Copenhagen 2016 competition. The study was approved by the Science ethical committee of the greater region of Copenhagen(H-15017269) and adhered to the Principles of the Helsinki declaration. Subjects received written and oral information about possible risks associated with the study before they volunteered to participate and signed a written informed consent form. The study meets the ethical standards of the International Journal of Sports Medicine[12].

Participants

Sixty-four healthy male volunteers were included in the study. They were told to refrain from vigorous exercise on the day before the test and eat their habitual mixed macronutrient diet. On the test day participants were questioned regarding their nutrition and exercise on the day before the test. Participants reported to the laboratory after an overnight fast no later than 11 am. All participants were fasted between 9–13 h. All participants reported to comply with the pre-visit standards. Male athletes of all levels were recruited within the age range from 20 to 50 yrs, through the official Ironman Copenhagen social media platform.

Experimental design

Body composition was determined using dual-energy X-ray absorptiometry (Lunar Prodigy Advance; Lunar, Madison, WI, USA). Participants were allowed water consumption in the morning and were encouraged to empty their bladder before the DXA-scan. Before the fatmax test a 12 mL resting venous blood sample was collected and immediately centrifuged at 4000 g for 10 min at 4 °C, and stored at −80 °C for later analysis (Centrifuge Hettich Universal 30 RF. Hettich Tuttlingen, Germany). For each blood sample; plasma glucose, lactate, glycerol, and FFA (EDTA, Vacutainer BD, New Jersey, United States) was analysed using a COBAS 6000 analyser 501C (Roche, Germany).

Before the the cycle ergometer (Monark 839E, Varberg, Sweden) exercise test, participants rested in the sitting position 5 min on the bike. Subsequently, the subjects performed a graded protocol commencing at 60 W followed by 35 W increases every 3 min using the last 90 s VO2 and VCO2 to compute a 3rd degree polynomial regression curve fit.This exercise protocol was adapted from Achten et al. (GE35/3protocol)[1]. When subjects reached a respiratory exchange ratio (RER) > 1.0 increments of 35 watts were induced every minute until voluntary exhaustion. VO2peak values were accepted when a levelling off or a decline in VO2 was reached despite increasing workloads and an RER above 1.15. Pulmonary VO2 and VCO2 were measured by the automated online system (Oxycon Pro system, Jaeger, Würzburg, Germany). The gas analyzers were calibrated with a 4.95 % CO2–95.05 % N2 gas mixture Substrate oxidation was calculated using the equation of Frayn with the assumption that urinary nitrogen excretion rate was negligible [9]:

Fat oxidation (g/min) = $(1.67 \times VO_2)$-$(1.67 \times VCO_2)$

Competition day

On the 21st of August, the 2016 Ironman Copenhagen started at 07.00 am and 2841 athletes swam 3.8 km in the artificial lagoon of Amager Strandpark, cycled a 180 km ride, consisting of two 90 km loops around the northern part of Zealand and finally ran 42.2 km in the centre of Copenhagen. The morning was cold and foggy with an air temperature of 11 degrees Celsius and the water temperature 18 degrees Celsius. The temperature increased throughout the day peaking at 24 degrees Celsius in the afternoon. The day was dry without rain and a humidity of 82 %, a slight breeze of average 3 m/s coming from south throughout the day.

Statistics

A Pearsons correlation matrix was computed including all relevant physiological variables regarding the primary outcome, race time. Simple bivariate regression analyses were carried out with race times and MFO as the dependent variables and MFO, VO2peak (relative to body mass), body fat percentage, maximal absolute VO2, plasma FFA and lactate as the independent variables.

Multiple regression analyses were performed with individual race time (min) and MFO (g/min) as the dependent variables. All significant independent variables from the bivariate analysis were entered into the multiple regression analysis. No independent variables were correlated and when an independent variable did not add to the prediction of the model, it was removed. A probability value of < 0.05 was accepted as significant and all data are presented as the mean ± SEM (▶ **Table 3**).

Sigmaplot. 13.0 (Systat Software, San Jose, CA, US) was used to calculate MFO and fatmax, which was done by constructing 3rd-degree polynomial regression with the best possible fit to the measured values. GraphPad Prism 6 (GraphPad Software, La Jolla, CA, US) was used to construct figures.

Results

Athletes

Sixty-four healthy Ironman athletes with a wide range of triathlon abilities were recruited for the study. Three participants were un-

▶ **Table 1** Performance times of 61 Ironman athletes.

Variable	Mean	Range
Race time	657.4 (10:57:24)	543.6–856.3 (09:03:34–14:16:37)
Swim time	74.4 (01:14:24)	48.33–109.43 (00:48:20–01:49:26)
Transition 1	5.9 (00:05:54)	2.3–9.5 (00:02:17–00:09:28)
Bike time	315.6 (05:15:35)	280.3–364.5 (04:40:15–06:04:29)
Transition 2	4.4 (00:04:24)	1.9–13.4 (00:01:52–00:13:26)
Marathon time	257.2 (04:17:12)	205.2–407.1 (03:25:11–06:47:06)
Performance times are given as minutes and (hours:minutes:seconds).		

able to complete the Ironman-distance triathlon, and their data are not included. The athletes' paticipating in this study were 35 ± 1 yrs. (range 23–47) old with a BMI of 23.6 ± 0.3 kg/m² (range 20.0–30.1), a body fat percentage of 16.7 ± 0.7 % (range 8.4–30.7) and a VO_2peak of 58.7 ± 0.7 ml/min/kg (range 43.9–72.5). At rest and overnight fasted mean plasma glucose concentration was 5.3 ± 0.0 mmol/L with a range of 4.7 to 6.1 mmol/L, mean plasma lactate concentration was 0.87 ± 0.03 mmol/L (range 0.4–1.6 mmol/L) and plasma glycerol and FFA concentrations were 68.2 ± 3.3 and 361 ± 25 µmol/L (range 36–155 and 114–995, respectively).

The mean race time for the 3.8 km swim, 180 km bikeride, and 42.2 km run was 657.4 min (10 h 57 min and 24 s) with a broad range between the fastest and slowest study participant (▶ **Table 1**). The mean swim time of 01:14:24 (hours:min:sec), bike time of 05:15:35 and runtime of 04:17:12 were 11.3, 48 and 39.1 % of total race time, respectively (▶ **Table 1**). Transition times were only 1.6 % of mean race time. The average performance times for all male participants in the Copenhagen Ironman event both overall and divided by discipline were approximately 5 % slower than compared to the study participants (▶ **Table 2**).

Fat oxidation rates

Mean maximal fat oxidation rate was 0.60 ± 0.02 g/min ranging between 0.34 and 1.00 g/min. When normalized to lean body mass MFO/LBM was on average 9.05 ± 0.27 mg/min/kg/LBM and the participants exhibited an extensive range from 5.04 to 14.61 mg/min/kg/LBM. The relative intensity that elicited the highest calculated rate of fat oxidation (FATMAX) was 45.0 ± 0.7 % VO_2peak ranging from 33 to 60 % VO_2peak.

Determinants of Race time and MFO

A pearsons correlation matrix were constructed including all relevant physiological variables and individual discipline times and race time. Age (yrs) were not associated with MFO but negatively associated with VO_2peak (ml/min/kg) and overall race time (▶ **Table 4**).

Simple bivariate analyses revealed significant negative correlations between race time and MFO ($r^2 = 0.12$, $p < 0.005$) (▶ **Fig. 1a**) and VO_2peak ($r^2 = 0.45$, $p < 0.0001$) (▶ **Fig. 1b**) and a positive correlation between race time and Body fat percentage ($r^2 = 0.27$, $p < 0.0001$) (▶ **Fig. 1c**). If performance times above 12 h are left out of the analysis the correlation between race time and MFO is ($r^2 = 0.27$, $p < 0.0001$, data not shown). Body weight, lean body mass or leg lean body mass were not related to race time (data not shown).

When MFO is applied as the dependent variable MFO and VO_2peak was not correlated, but a positive correlation was observed with VO_2 ($r^2 = 0.13$, $p < 0.005$) and fasting plasma FFA concentrations ($r^2 = 0.09$, $p < 0.05$). Fasting plasma lactate concentrations and MFO was negatively correlated ($r^2 = 0.12$, $p < 0.01$) (▶ **Fig. 2**). MFO was positively correlated also to FATMAX, lean body mass and fasting plasma glycerol concentrations, but not body fat percentage and fasting plasma glucose concentration (data not shown). FATmax and racetime were not correlated.

Multiple regression analysis

When the significant variables from the bivariate regression analyses were entered into the multiple regression models, VO_2peak and MFO together explained 50 % of the variation observed in race time among the 61 Ironman athletes (adj $R^2 = 0.50$, $p < 0.001$). VO_2, fasting plasma lactate concentration and fasting plasma FFA concentration together accounted for 23 % of the variation in MFO (adj $R^2 = 0.23$, $p < 0.001$).

Discussion

The main findings of this study were that 50 % of the variation in ironman triathlon race time could be explained by peak oxygen uptake and maximal fat oxidation and maximal fat oxidation rate independently contributed significantly to explain ultra-endurance performance.

In the present study, we found a significant association between ironman triathlon racetime and both VO_2peak and MFO in a relatively large and heterogenous group of male ironman athletes all performing the same triathlon. Furthermore, we found that regression analysis revealed that VO_2peak and MFO could explain 50 % of the variation observed overall in ironman triathlon race time. Based on the bivariate regression VO_2peak exhibited a more robust ($r^2 = 0.45$) association with ironman triathlon race time than MFO ($r^2 = 0.12$). A number of studies have demonstrated a direct coupling between VO_2peak and olympic distance triathlon performance, but to the best of our knowledge, the association between peak or maximal oxygen uptake in ironman athletes and the relation to Ironman triathlon performance has not previously been reported [5, 8, 30]. However, several reviews Laursen and Rhodes [19] and recently Knechtle et al. [16] implies that the relationship is obviously there, but also that predicting Ironman triathlon performance and particular elite athlete ironman triathlon performance is a complex issue. Importantly, we note that Laursen and colleagues report peak oxygen uptakes in 24 well-trained ultra-endur-

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ance triathletes measured on cycle-ergometer of 4.72 ± 0.42 L/min, which is similar to the 4.52 ± 0.67 L/min observed in this study [18]. When the data is expressed relative to bodymass mean VO_2peak was 63.9 ml/min/kg and this is 9 % above (58.7 ± 0.7 ml/min/kg) what was measured in the athletes in our study. Although comparison of ironman race times on different courses should be done with caution as small variations in the nature of the course and weather can account for large variations in ironman triathlon racetime, there was also a similar difference (7 %) in the average personal best ironman triathlon race time (10:12 ± 0:32 h: min) and (10:57 ± 0:08 h: min), reported in Laursen et al. 2005 and our study, respectively. Overall we think that our sample of 61 Ironman triathlon athletes are representative of the recreational active athletes that endevour into doing ironman triathlon competitions.

Endurance training increases fat oxidation during exercise at the same absolute workload [23], and yet there is to date no real strong evidence that increased fat oxidation is directly coupled to endurance or ultra-endurance performance (>4 h. exercise). Fat oxidation rates have been studied extensively in well trained and ultra endurance trained athletes, both in fasted state acustomed to a habitual diet [2, 3, 24] and in the fasted state acustomed to high fat diets [25, 29]. However, an association between individual rates of maximal fat oxidation and ultra-endurance performance has to our knowlegde never been reported. In this study, we show an association between MFO and ultra endurance performance independent of the relation between peak oxygen uptake (ml/min/kg) and ironman triathlon performance. Interestingly, we found no association between VO_2peak and MFO despite the relatively large variation in VO_2peak in the 61 subjects included in our study. This is contrast to the study by Venables et al. where a relationship

between VO_2max and MFO was observed in a very heterogenous group of 300 men and women [28]. In a prior study, we also observed a relationship between VO_2peak and MFO when comparing untrained and very trained young men, and it may suggest that this association is only present when comparing heterogenous groups [24, 26, 28].

In the present study there was no association between FATMAX and overall performance (race time) despite a robust positive association between MFO and FATMAX. In line with our finding Jeukendrup and co-workers previously found that VO_2max and FATMAX were not related in a group of well-trained athletes [2]. Ironman athletes are expected to follow nutrional guidelines and competitions are therefore performed in the fed state, which is in contrast to our tests that were performed in the fasted state. It has recently been shown that carbohydrate ingestion during endurance exercise modulates liver but not muscle glycogen concentration [11], and in a prior study we demonstrated that muscle glycogen was markedly lowered after very prolonged ultraendurance exercise [13]It is likely that the difference in feeding and probably lowered muscle glycogen, at least partly, explain the lack of association between FATMAX and race performance time.

In order to establish which, varibles predicts the relatively large variation in MFO (range: 0.34–1.00 g/min) a multiple regression analysis was performed with VO_2 at FATMAX, fasting plasma lactate and FFA concentrations as independent variables. These independent variables were the strongest predictors of the observed variation in MFO, but in total, they only account for 23 % of the variation in MFO. This is in line with Venables and colleagues where FFM (Fat Free Mass), SRPAL (self-reported physical activity level), Gender, VO_2max and FM (Fatmass) could only explain 36 % of the variation in MFO (range 0.18–1.01 g/min) in a fairly large sample of 300 individuals. In the present study, there was no association between FM and MFO, but we did find an association between lean body mass (LBM) and MFO ($r^2 = 0.12$, $p < 0.005$).
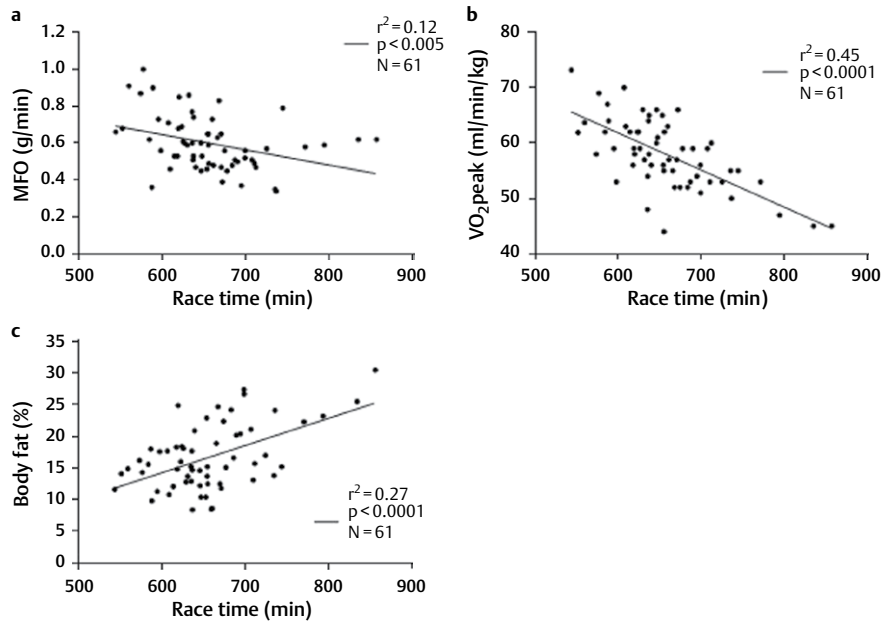
The association between plasma concentration of FFA and lactate with MFO has previously been demonstrated by Goedecke and collegaues, and it support the notion that high plasma FFA availability and low resting lactate concentrations results in elevated fat oxidation i.e low RER [10, 27]

In an attempt to avoid an effect of age we recruitted subjects from a relatively narrow range of age (23–47 yrs). In our laboratory we have previously seen that age negatively influences MFO in both endurance trained and untrained subjects (unpublished data),

▶ **Table 2** Performance times of all male participants in the competition (n = 1931).

Variable	Mean	% diff.
Race time	695.1 (11:35:05)	5.6
Swim time	76.6 (01:16:36)	2.9
Transition 1	7.5 (00:07:30)	23.8
Bike time	334.0 (05:34:00)	5.7
Transition 2	5.5 (00:05:30)	22.2
Marathon time	271.5 (04:31:30)	5.4
Performance times in minutes (hours:minutes:seconds). Percentage difference from the 61 study participants		

▶ **Table 3** Individual bivariate and multiple regression analyses.

Dependent variable	Independent variables	Correlations			Coefficients
		R	R²	Adj. R²	P-value
Race time	VO₂peak (mL/min/kg)	0.67	0.45	0.44	<0.001
	MFO (g/min)	0.35	0.12	0.11	<0.01
All significant variables		0.72	0.52	0.50	<0.001
MFO	VO₂ (mL/min)	0.36	0.13	0.11	<0.01
	Fasting plasma lactate (mmol/L)	0.35	0.12	0.10	<0.01
	Fasting plasma FFA (µmol/L)	0.28	0.08	0.06	<0.05
All significant variables		0.52	0.27	0.23	<0.001
n = 61					

▲ **Table 4** Peasons correlation matrix.

Pearsons correlation matrix		Fatmax (% VO$_2$peak)	MFO (g/min)	VO$_2$peak (ml/min/kg)	VO$_2$ (ml/min)	Lean body mass (kg)	Body fat (%)	Age (yrs)	Swim time (min)	Bike time (min)	Run time (min)	Race time (min)
Fatmax (% VO$_2$peak)	P-value		<0.001	ns	ns	<0.05	ns	ns	ns	ns	ns	ns
	r		0.67	-0.12	0.08	0.26	-0.07	0.16	0.09	-0.10	-0.13	-0.10
MFO (g/min)	P-value	<0.001		ns	<0.01	<0.01	ns	ns	<0.05	<0.05	<0.05	<0.01
	r	0.67		0.12	0.36	0.34	-0.09	0.09	-0.29	-0.32	-0.28	-0.35
VO$_2$peak (ml/min/kg)	P-value	ns	ns		<0.001	ns	<0.001	<0.05	<0.01	<0.001	<0.001	<0.001
	r	-0.12	0.12		0.50	-0.16	-0.56	-0.32	-0.35	-0.55	-0.64	-0.67
VO$_2$ (ml/min)	P-value	ns	<0.01	<0.001		<0.001	<0.001	ns	<0.001	<0.001	<0.001	<0.001
	r	0.08	0.36	0.50		0.65	-0.60	-0.10	-0.44	-0.54	-0.34	-0.49
Lean bodymass (kg)	P-value	<0.05	<0.01	ns	<0.001		ns	ns	=0.07	<0.05	ns	ns
	r	0.26	0.34	-0.16	0.65		-0.04	0.14	-0.23	-0.25	-0.02	-0.15
Body fat (%)	P-value	ns	ns	<0.001	<0.001	ns		ns	ns	<0.01	<0.001	<0.001
	r	-0.07	-0.09	-0.56	-0.60	-0.04		0.16	0.10	0.35	0.59	0.52
Age (yrs)	P-value	ns	ns	<0.05	ns	ns	ns		ns	<0.05	<0.001	<0.01
	r	0.16	0.09	-0.32	-0.10	0.14	0.16		0.19	0.31	0.34	0.37
Swim time (min)	P-value	ns	<0.05	<0.01	<0.001	=0.07	ns	ns		<0.001	<0.001	<0.001
	r	0.09	-0.29	-0.35	-0.44	-0.23	0.10	0.19		0.53	0.59	0.61
Bike time (min)	P-value	ns	<0.05	<0.001	<0.001	<0.05	<0.01	<0.05	<0.001		<0.001	<0.001
	r	-0.10	-0.32	-0.55	-0.54	-0.25	0.35	0.31	0.53		0.58	0.82
Run time (min)	P-value	ns	<0.05	<0.001	<0.001	ns	<0.001	<0.001	<0.001	<0.001		<0.001
	r	-0.13	-0.28	-0.64	-0.34	-0.02	0.59	0.34	0.59	0.58		0.92
Race time (min)	P-value	ns	<0.01	<0.001	<0.001	ns	<0.001	<0.01	<0.001	<0.001	<0.001	
	r	-0.10	-0.35	-0.67	-0.49	-0.15	0.52	0.37	0.61	0.82	0.92	

N = 61, ns = Not significant, MFO = Maximal fsat oxidation rate

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▶ **Fig. 1** Linear regression plot of MFO **a**, VO₂peak **b**, and body fat percentage **c** with race time as the dependent variable in 61 male ironman triathlon participants.
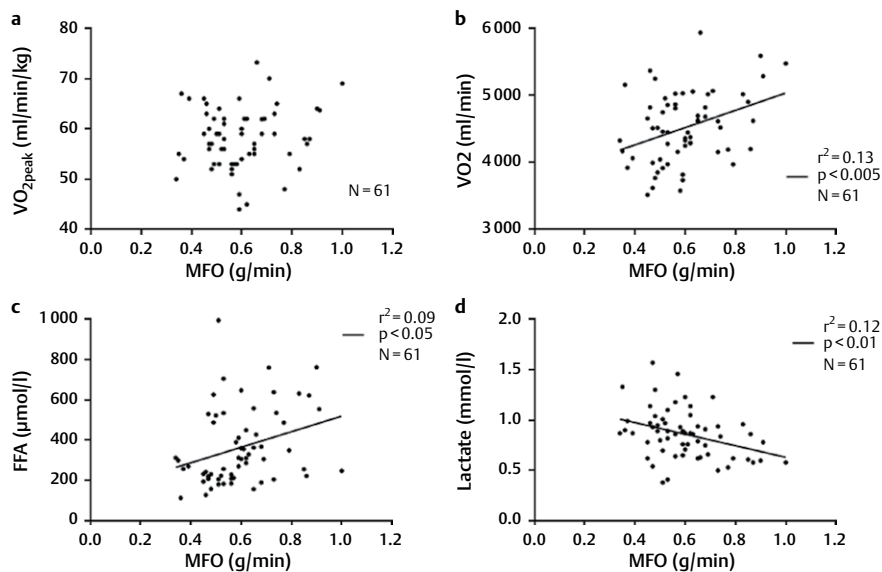
▶ **Fig. 2** Scatterplot of correlation between MFO and VO₂peak **a** and linear regression plots of VO₂ **b**, fasting plasma FFA concentrations **c** and fasting plasma lactate concentrations **d** with MFO as the dependent variable in 61 male ironman triathlon participants.

but as we observed no association between age and MFO in the present study, we succeeded in minimizing a possible influence of age (data not shown). The anthropometric characteristics of the athletes included in this study are in accordance with previously reported findings in 27 nonprofessional male ironman athletes participating in the 2007 Ironman Switzerland and also 83 recreation-

al male ironman athletes participating in the 2009 Ironman Switzerland. with an age of 30.3 ± 9.1 (SD) and 41.5 ± 8.9 yrs. (SD) and a body fat % of 14.4 ± 4.8 (SD) and 15.7 ± 4.6 (SD), respectively [14, 15]. In the present study, we did not measure substrate utilization and contribution of exogenous and endogenous substrate during the ultra-endurace event, but we speculate that a higher ca-

pacity for fat oxidation during exercise would spare muscle and liver glycogen and thus over time allow maintenance of a higher overall exercise intensity. Further research is needed to identify the cellular mechanisms that may explain this observation.

It is a limitation to the present study that we did not control for variables such as prerace ironman triathlon experience, previous training, and cycling aerodynamics, running mechanics and energy and macronutrient intake during the triathlon competition. It is also acknowledged that, testing in the fasted state and test modality i. e. cycling vs. running may also pose a further limitation to the study design as the Ironman triathlon is most likely performed in fed state and incorporate three different exercise modalities. Although these variables could influence the race time [15], we aimed for a simple study design and only investigated to what extent VO$_2$peak, MFO, plasma metabolites and subject characteristics could predict the differences in race time. Thus, despite not controlling all these factors the associations between ironman triathlon race time and MFO and VO$_2$peak still emerged as significant.

Conclusion

In accordance with our hypothesis, we show that maximal fat oxidation rate is associated with overall Ironman performance independent of peak oxygen capacity, where the latter was the strongest individual predictor of overall Ironman performance. Furthermore, we demonstrate that 50 % of the variation in Ironman triathlon race time can be explained by peak oxygen uptake and maximal fat oxidation. Future research should aim to investigate the factors that influence MFO and how this mechanistically may enhance ultra-endurance performance.

Author contribution

JF, SDV, SL, FD and JWH contributed to data acquisition, analyses and interpretation. JF and JWH wrote the manuscript. All authors revised the manuscript and approved the final article.

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Conflict of Interest

The authors declare no conflict of interests.

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