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LETTER TO THE EDITOR

Methodological issues related to maximal fat oxidation rate during exercise

Comment on: Change in maximal fat oxidation in response to different regimes of periodized high-intensity interval training (HIIT)

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Dear Editor,

We read the study by Astorino et al. (2018) with interest, which examined the effects of three different high-intensity interval training (HIIT) programs on maximal fat oxidation (MFO) and on the exercise intensity that elicits MFO (FATmax) during a graded exercise test in young active adults after 6 weeks. They found no differences in MFO and FATmax when comparing the HIIT groups with a non-exercising control group. These findings do not concur with other studies that reported an increase in MFO and FATmax (1) in young active adults after 6 weeks of HIIT (Perry et al. 2008) (2) in healthy middle-aged adults after 12 weeks of HIIT (Bagley et al. 2016), and (3) in young active women after 2 weeks of HIIT (Talanian et al. 2007) (see Table 1). Astorino et al. (2018) argued that the lack of differences in MFO and FATmax could explain (1) the marked interindividual variability obtained in the MFO and FATmax values (~ 25% coefficient of variance), and (2) the overall HIIT volume performed.

Astorino et al. (2018) included a non-exercising control group, which certainly is a strength, yet the daily amount of physical activity of this group was not well controlled during the intervention program. Considering that they were physically active (> 150 min/week) and that some of them were CrossFit exercisers or recreational endurance athletes before the start of the study, an objective physical activity measurement by accelerometry should have been considered. In addition, it is important to note that they did not control the menstrual cycle variation during the test protocol, which is a well-accepted factor that modifies MFO and FATmax during exercise (Purdom et al. 2018). These factors, may partially explain the lack of differences across groups in MFO and FATmax in the study by Astorino et al. (2018).

In addition there is a number of other factors that could help to better understand why one study did not find differences in MFO and FATmax (Astorino et al. 2018), while others did (Bagley et al. 2016; Perry et al. 2008; Talanian et al. 2007):

1. Besides the HIIT volume as a factor that may affect MFO and FATmax changes after a HIIT intervention program (Astorino et al. 2018), other exercise training variables, such as the training frequency, the training intensity, or the HIIT modality, should be taken into account when different studies are compared (see Table 1).
2. Training status: it is well known that trained individuals have a greater ability to oxidize fat at higher exercise intensities (Purdom et al. 2018). MFO has been positively associated with improvements in respiratory capacity (measured by the VO_2max) and also with higher intramuscular triglyceride concentrations, fatty acids plasma availability and transport, and mitochondrial density and activity (Purdom et al. 2018). All of these physiological processes are related to chronic adaptations induced by HIIT. However, there is still controversy when comparing MFO and FATmax in individuals with different training status, or even with

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Table 1 Descriptive and methodological characteristics of all studies compared

	Astorino et al. (2018)	Perry et al. (2008)	Bagley et al. (2016)	Talanian et al. (2007)
Sex	Men/women (*N* = 34/37)	Men/women (*N* = 5/3)	Men/women (*N* = 24/17)	Women (*N* = 8)
Age (years)	23.3 ± 4.6	24.0 ± 1.0	39.0 ± 2.0	22.0 ± 1.0
Weight (kg)	69.6 ± 10.0	72.7 ± 4.0	71.3 ± 2.4	65.0 ± 2.2
Height (cm)	171.0 ± 1.4	179.0 ± 4.0	172.4 ± 9.7	Not reported
VO_2max (ml/kg/min)	40.2 ± 5.5	45.3 ± 3.3	38.2 ± 2.3	36.3 ± 3.7
Training status	Active	Active	Not reported	Active
Training program duration (week [sessions])	6 [20]	6 [18]	12 [36]	2 [7]
Training program volume (min)	~475	~720	~360	~280
Training program intensity	90% of VO_2max	90% of VO_2max	175% of VO_2max	90% of VO_2max
Session duration (min)	23–35	40	10	40
Test protocol modality	Incremental	Constant	Incremental	Incremental
Total duration time (min)	~25	60	~20	60
Stage duration (min)	3	60	3	60
Test ergometer	Cycle ergometer	Cycle ergometer	Cycle ergometer	Cycle ergometer
Test intensities imposed	Increment of 20W until RER = 1	60% VO_2max	Increment of 50W (men) or 30W (women) until RER = 1	~60% VO_2max
Fasting time (hours)	Not reported	2	12	3–4
Diet standardization prior to the test	Controlled (4 days before)	Controlled (2 days before)	Not controlled	Controlled (1 day before)
Menstrual cycle variations	Not controlled	Controlled (early to mid-follicular phase)	Not controlled	Not controlled
Gas analyzer	ParvoMedics TrueOne, Sandy, UT	Vmax 229, Sensormedic, Yorba Linda, CA	Metalyzer 3B, Cortex, Leipzig, Germany	Vmax 229, Sensormedic, Yorba Linda, CA
Time interval selected	Last 60 s of each stage	15–20/35–40/55–60 min	Not reported	13–17/28–32/43–47/55–59 min
Stoichiometric equation	Frayn	Peronnet	Frayn	Peronnet
FATmax determination method	Not reported	Not reported	Best-fit polynomial curve	Not reported

Data are presented as mean ± standard deviation

VO_2max Maximal oxygen uptake, *RER* Respiratory exchange ratio, *FATmax* the exercise intensity that elicits maximal fat oxidation

untrained individuals with different body compositions (Purdom et al. 2018). Astorino et al. (2018) compared their results with other studies that recruited participants with different VO_2max values (Bagley et al. 2016; Perry et al. 2008; Talanian et al. 2007) and, consequently, with different training status. Therefore, these comparisons should be considered cautiously.

3. Test protocol: although an incremental exercise test protocol to determine MFO was validated almost 20 years ago, several MFO test protocols have been described until the present time. The studies compared used different test protocol modalities [incremental (Astorino et al. 2018; Bagley et al. 2016) vs. constant (Perry et al. 2008; Talanian et al. 2007)], imposed different intensities [increment of 20W (Astorino et al. 2018) vs. 50W in men and 30W in women (Bagley et al. 2016) vs. a constant load of 60% of VO_2max], and had different total duration [20–60 min], different fasting times [2–12 h], and also different diet standardizations prior to the tests [not controlled (Bagley et al. 2016) vs. controlled 24-h prior to the test (Talanian et al. 2007) vs. controlled 48-h prior to the test (Perry et al. 2008) vs. controlled 96-h prior to the test (Astorino et al. 2018)]. It is important to note that only Perry et al. (2008) controlled the menstrual cycle variation during the test protocol.
4. Data analysis method: there are currently no widely accepted standardized protocols established for MFO and FATmax data analysis during exercise. In this case, the studies analyzed different time intervals and used two different stoichiometric equations [Frayn (Astorino et al. 2018; Bagley et al. 2016) vs. Peronnet (Perry et al. 2008; Talanian et al. 2007)]. Moreover, it is still unclear whether the use of different time intervals, stoichiometric equations, or also different FATmax determination

methods (i.e., measures values method vs. polynomial curves vs. sinusoidal model) could affect MFO and FATmax values during a graded exercise test.

In summary, we believe that, in addition to the marked interindividual variability in MFO and FATmax described by Astorino et al. (2018), there are other important aspects that must be taken into account when the results of different studies are analyzed and compared. More investigations are required to elucidate which is the best approach to measure and analyze MFO and FATmax during exercise.

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