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




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Relationship of Testosterone, LH, Estradiol, IGF-1, and SHBG with Physical Performance of Master Athletes

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ABSTRACT

Purpose: The aim of this study was to investigate and compare the levels of luteinizing hormone (LH), testosterone (T), estradiol (ES), sex hormone-binding globulin (SHBG), and insulin-like growth factor 1 (IGF-1) in master sprint (MS) and master endurance (ME) athletes. Additionally, the possible associations between these hormones, body composition, and lipid profile with athletic performance (% of performance in relation to the current world record) were analyzed. **Materials and Methods:** The participants were all men: (i) 34 MS (51.0 ± 6.8 years); and (ii) 32 ME (51.7 ± 9.4 years). Student's t-tests for independent samples were performed to compare all variables between groups. **Results:** MS had a significantly higher ($p = .008$) average IGF-1 (154.78 ± 29.85 ng/mL) when compared to ME (129.92 ± 25.48 ng/mL). Performance was significantly correlated with IGF-1 ($r = 0.424$). The MS group had a moderately lower body fat than ME athletes (MS 12.54 ± 4.07 vs. ME 14.60 ± 4.12; $p = .078$; $d = 0.503$). **Conclusions:** Thus, strength/power training exercise/sport seems to be more beneficial for obtaining a higher IGF-1 compared to aerobic/distance exercise/sport. In addition, LH, T, ES, and SHBG were similar between the two groups of athletes and were comparable to the reference values of younger adults.

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The number of people over 60 years old and the average life expectancy of the population is increasing (United Nations DoE, Social Affairs PD, 2019). Moreover, the number of senior adults participating in competitive sporting events has risen dramatically (Donato et al., 2002; Zaryski & Smith, 2005). Adults competing in nonprofessional but competitive sports events are referred to as master athletes, and they range between 35 and 110 years old (Tanaka & Seals, 2008). Competitions for master athletes are organized at regional, national, and world levels and are highly technical (Concannon et al., 2012). Studies of ranked master athletes are important because physiology and performance related to many decades of training by different events and sports can be observed (Nikolaidis et al., 2017). Thus, identifying characteristics specific to athletic performance among master athletes compared to younger athletes and understanding how exercise training mitigates the decrease in physical functioning over the lifespan are interesting and worthwhile topics of research. Also, master athletes have a better quality of life and a lower risk of chronic diseases, providing a potentially useful subgroup for comparisons to those succumbing to such morbidity and mortality (Rebello-Marques et al., 2018).

Among the different factors of human performance (especially athletic), the endocrine function is crucial. Men's sex-specific hormones are secreted from the pituitary gland and gonads: luteinizing hormone (LH), testosterone (T), and

estradiol (ES) from the conversion of T to ES (Kaufman & Vermeulen, 2005). In addition to proteins, sex hormone-binding globulin (SHBG) is mainly responsible for binding T, leaving it inactive. Finally, insulin-like growth factor 1 (IGF-1), molecularly similar to insulin, is produced from growth hormone (GH) signaling stimulus and leads to tissue growth, mainly skeletal muscle. These hormones (LH, T, ES, and IGF-1) have fundamental functions in macronutrient metabolism, maintenance of muscle and bone mass, and behavior and cognition (Joshi & Parle, 2006; Maggio et al., 2013). Such hormones provide the necessary support for better sports performance, including at the master level. Conversely, low secretion of these hormones is associated with several age-related conditions, such as loss of muscle and bone mass, an increased risk of atherosclerosis, and insulin resistance (1), which could all negatively affect physical performance. However, the impact of the hormonal profile of master athletes on their performance is uncertain, especially comparing endurance athletes to power athletes, such as sprinters.

Among master athletes, physical and physiological differences occur due to their sport specialization. For instance, master sprint (MS) or power athletes (i.e., 60, 100, 200, and 400 m and 110 and 400 m hurdles events) have higher body mass and muscle mass than master endurance (ME) athletes (i.e., 10000 m, half-marathon, and marathon). These two types of athletes have different performance goals, and therefore,

different training regimens (Kusy & Zielinski, 2015). This typically leads to distinct physical and physiological adaptations over years of training.

Studies of ME athletes (Ari et al., 2004; Tissandier et al., 2001) show that they have more testosterone than age-matched controls. Hayes and Grace evaluated 20 male ME athletes (60.4 ± 4.7 years of age) and found that the testosterone level was higher in master athletes (493.8 ± 180.3 pmol/L) when compared to the age-matched control group, suggesting that exercise training is essential in improving the physiological and metabolic profile of older individuals (Hayes et al., 2013). Additionally, recent research on master athletes shows that their performance is improving (Lepers & Stapley, 2016), particularly among those athletes over 60 years of age (Lepers & Cattagni, 2012). This may be due to an artifact of greater participation of master athletes in competitive events, increasing the chance of better performances (Hunter & Stevens, 2013). However, to date, no study has assessed and compared the hormonal profile and athletic performance between ME and MS athletes. Thus, the aim of this study was to investigate and compare the levels of the LH, T, ES, SHBG, and IGF-1 in MS and ME athletes and the relationships between those hormones with athletic performance and body composition.

Methods

This observational study was approved by the Ethics Research Committee for humans studies at the Catholic University of Brasilia (protocol number: 3,779,535), and was conducted according to the standards of the Helsinki declaration.

Each participant volunteered and provided written informed consent after all procedures had been fully explained and before participation in the study. Master athletes were recruited at a national and an international track and field events and from personal recommendations from athletes. The inclusion criteria were: men between 40 and 75 years of age; no history of inflammatory or metabolic diseases or cancer; nonsmoker; no regular use of drugs, including hormone replacement therapies. The selected participants had at least 15 years of regular and competitive practice in sprint (i.e. 100-400 m dash, 110 m hurdles, 400 m hurdles, long jumpers, high jumpers ...) or endurance events (i.e. 3,000 to 10,000 meter runners, and marathoners); and have participated either in international or national track and field meetings at the current year of data collection. These athletes compete according to their age group, with an interval of 5 years in each category (M35–39, M40–44, M45–49 ... M95–99, M100⁺), to promote a fair competition among participants. Master athletics competitions are divided into local, national, regional, continental, and world levels (ATHLETICS, 2021a; b).

Participants completed a questionnaire with personal and training information, underwent an anthropometric evaluation, and had their blood collected. Before venipuncture, subjects were required to be fasting for 8 hours and had to be at complete rest with no physical exercise in the last 8 hours. Two samples of ~4 mL each of blood were collected using Vacutainer tubes with and without EDTA. One tube of blood was centrifuged for serum isolation and triglycerides, high-density lipoprotein (HDL-c), total cholesterol, and glycemia

using commercial kits (Labtest[®], Minas Gerais, Brazil), according to the instructions of the manufacturer. Low-density lipoprotein was determined using the Friedewald formula (Friedewald et al., 1972). A single evaluator collected all seven skinfold measurements, considering a mean of three measurements for each skinfold, using a Lange[®] caliper (Cambridge Scientific Instruments, Maryland, USA). The relative body fat was estimated according to the Jackson and Pollock protocol (Jackson & Pollock, 1978). Body density was then calculated and converted to body fat percentage utilizing an equation described elsewhere (Siri, 1961).

Venous blood was collected and used for chemiluminescence hormonal analyses (LH, T, ES, SHBG, and IGF-1) at an outsourced reference laboratory. LH, T, ES, and SHBG analyses were measured using the Atellica[®] - Siemens[®] automatic immunoassay equipment. IGF-1 was measured using the Immulite[®] - Siemens[®] automated immunoassay equipment.

The level of performance of each athlete (% of performance in relation to the current world record) was calculated individually, considering each athlete's best performance in the current season, with the record of his event, in their corresponding age and gender groups.

For the statistical analysis, the Shapiro-Wilk test verified the normality of the data. The data were expressed as mean \pm standard deviation. Student's t-tests for independent samples were performed to compare all variables between MS and ME athletes. In addition, a moderate effect size (Cohen's $d = 0.7$) with a significance level of $\alpha = 0.05$ for the total sample in the study ($n = 66$) conferred 88% statistical power (Cohen, 2013). Pearson's correlation coefficients were applied to determine the associations between performance level, hormones, and body composition (percent body fat and body mass index [BMI]). A secondary analysis was performed stratifying the performance of all participants combined in tertiles, in which tertile 1 ($T1 \leq 81.1\%$), tertile 2 ($T2 81.2\% - 89.8\%$), and tertile 3 ($T3 \geq 89.9\%$) were defined. The objective was to compare "lower performance" (T1) with "higher performance" (T3), so T2 was excluded from the analyses. Thus, T1 and T3 were compared with Student's t-tests on body fat and hormonal parameters. The level of significance was set at $p < .05$. All analyses were performed using GraphPad Prism (v6.0), Gpower[®] (v3.1), and IBM SPSS Statistics 21 for Windows (IBM, Inc., Chicago, IL, USA).

Results

The participants were: (i) 34 MS athletes (51.0 ± 6.8 years old; specializing in 60, 100, 200, and 400 m and 110 and 400 m hurdles events) and (ii) 32 ME athletes (51.7 ± 9.4 years old; specializing in 10,000 m, half-marathon, and marathon events). For the total sample, participants averaged 25.0 ± 9.9 years of training, 9.3 ± 3.7 hours per week of training, and 8.3 ± 5.5 competitions per year. The best self-reported performance by the participants ranged from 68% to 99% in relation to their respective age group world record (5-year intervals) registered at the World Masters Athletics (World Masters Athletics, 2019). In addition, every participant had reached the victor's podium at least once in either the Brazilian, South American or world championship.

Table 1. Age, body composition, training time, competitions per year, lipid profile, and blood glucose of master sprint and master endurance athletes. Data are expressed as mean \pm standard deviation with the corresponding *p* value and reference value, where necessary.

Variable	MS (<i>n</i> = 34)	ME (<i>n</i> = 32)	<i>p</i> value	Reference value
Age (years)	51.0 \pm 6.7	51.7 \pm 9.3	0.709	
Height (cm)	176 \pm 5	173 \pm 6	0.053	
Body mass (kg)	75.5 \pm 7.6	70.8 \pm 7.3	0.015	
BMI (kg/m (Donato et al., 2002)	23.5 \pm 4.6	23.5 \pm 2.2	0.205	
Lean body mass (%)	87.5 \pm 4.5	85.4 \pm 4.1	0.047	
Training time (years)	26.2 \pm 11.0	23.5 \pm 8.5	0.309	
Competition (years)	7.0 \pm 5.3	12.7 \pm 11.2	0.016	
Triglyceride (mg·dl ⁻¹)	75.1 \pm 32.3	100.7 \pm 40.2	0.015	<150
HDL-c (mg·dl ⁻¹)	68.8 \pm 28.9	74.9 \pm 36.8	0.484	>40
LDL-c (mg·dl ⁻¹)	105.1 \pm 46.7	111.4 \pm 53.7	0.671	<110
Total cholesterol (mg·dl ⁻¹)	192.2 \pm 43.5	206.2 \pm 46.1	0.256	<190
Blood glucose (mg·dl ⁻¹)	86.4 \pm 22.1	85.9 \pm 7.3	0.755	<100

Note: Values for lipid profile based on individuals with low cardiovascular risk.

The characteristics of each group are displayed in Table 1. MS had a higher body mass. Higher lean mass percentage. Lower triglycerides. And competed in fewer competitions per year when compared to ME. No other variables were significantly different between the two groups.

MS athletes had a significantly higher average IGF-1 (154.78 \pm 29.85 ng/mL) compared to ME athletes (129.92 \pm 25.48 ng/mL) with a large effect size (*p* = .008; *d* = 0.896) (Figure 1C). No significant differences were found for LH (MS 3.93 \pm 1.33 mIU/mL vs. ME 3.24 \pm 1.24 mIU/mL; *p* = .069; *d* = 0.533) (Figure 1D), T (MS 644.45 \pm 176.71 ng/dL vs. ME 717.22 \pm 156.38 ng/dL; *p* = .127; *d* = 0.436) (Figure 1A),

ES (MS 32.06 \pm 5.08 pg/mL vs. ME 36.80 \pm 10.70 pg/mL; *p* = .067; *d* = 0.533) (Figure 1B), and SHBG (MS 30.80 \pm 14.12 nmol/L vs. ME 26.45 \pm 12.48 nmol/L; *p* = .240; *d* = 0.326) (Figure 1E).

All participants were stratified together in tertiles according to individual performances, and those in T1 (\leq 81.10%) were compared to those in T3 (\geq 89.9%). T3 athletes had a higher IGF-1 (155.27 \pm 28.31 ng/mL) compared to T1 athletes (126.00 \pm 26.26 ng/mL; *p* = .029; *d* = 1.072) with a large effect size (Figure 2C). SHBG was also higher in T3 athletes (36.50 \pm 18.11 nmol/L) compared to T1 athletes (21.37 \pm 7.21 nmol/L; *p* = .024; *d* = 1.098) with a large effect size (Figure 2E). While

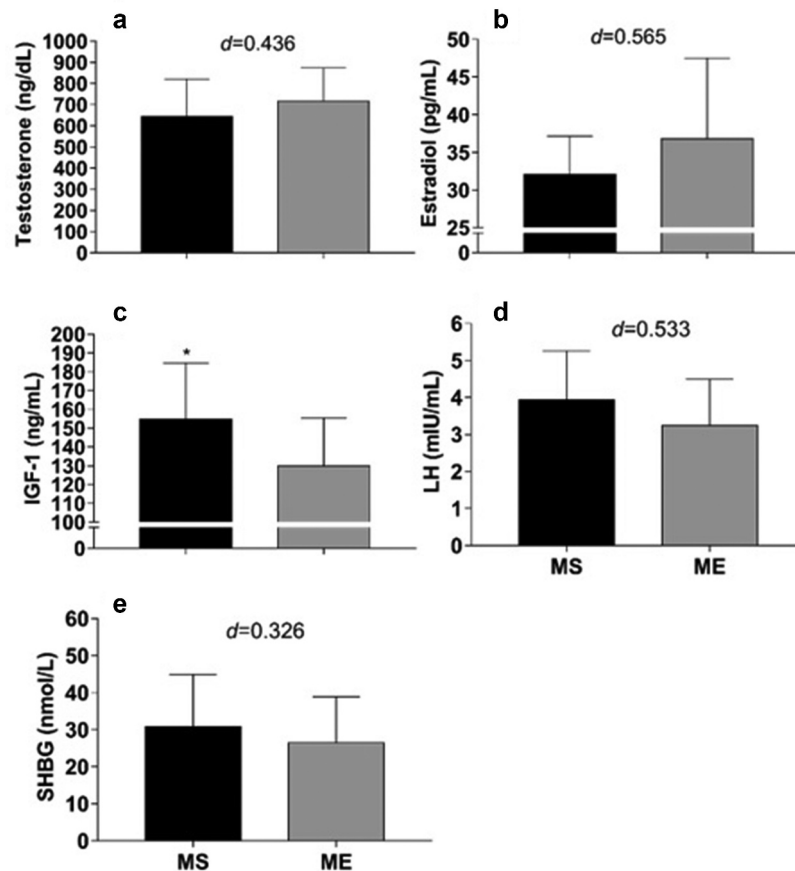


Figure 1. Comparisons of the hormonal profile between master sprint and master endurance athletes. **p* < .05.

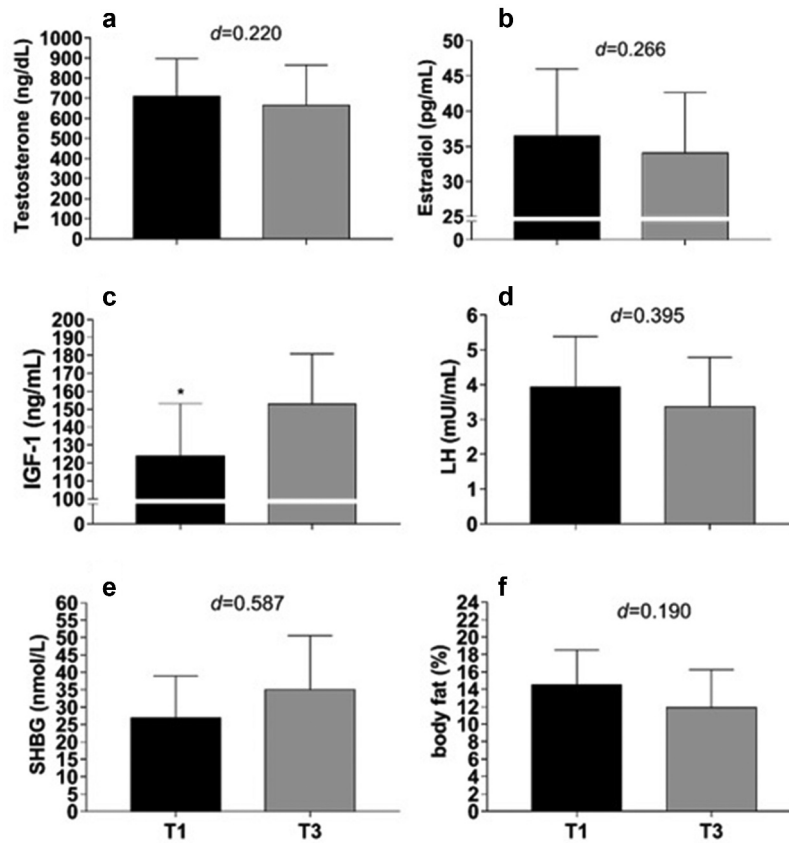


Figure 2. Comparisons of the hormonal profile between low-performance and high-performance master athletes. * $p < .05$.

non-significant, T3 athletes ($11.91 \pm 4.33\%$) had a lower body fat percentage compared to T1 athletes ($14.50 \pm 4.00\%$) with a moderate effect size ($p = .107$; $d = 0.619$) (Figure 2F). No statistical differences were found for T (T3 665.70 ± 199.94 ng/dL vs. T1 708.42 ± 188.03 ng/dL; $p = .612$; $d = 0.220$) (Figure 2A), LH (T3 3.36 ± 1.41 mIU/mL vs. T1 3.93 ± 1.45 mIU/mL; $p = .358$; $d = 0.395$) (Figure 2D), or ES (T3 34.05 ± 8.57 pg/mL vs. T1 36.47 ± 9.55 pg/mL; $p = .540$; $d = 0.266$) (Figure 2B).

Performance was significantly correlated with IGF-1 ($r = 0.424$; $p = .013$) (Figure 3). No correlations were found

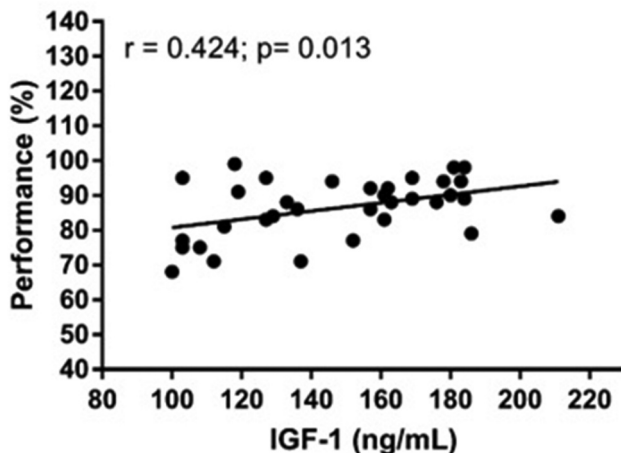


Figure 3. Correlation between performance and IGF-I.

between performance and T, ES, LH, SHBG, body composition, and lipid profile. No correlations were found between lipid profile and T, ES, LH, and SHBG.

Discussion

The present study analyzed and compared LH, T, ES, SHBG, IGF-1, body composition, and lipids between MS and ME athletes. Moreover, these variables were analyzed within the context of the performance level of participants. The main findings were that master athletes' specialization does not differentiate LH, T, ES, and SHBG. However, MS had higher lean mass, lower triglycerides, and higher IGF-1 compared to ME. In addition, athletes with higher individual performance had lower body fat percentage and a higher level of IGF-1 when compared to athletes stratified with low performance.

Competitive athletic practice throughout life seems to preserve the hormonal profile of master athletes, regardless of the type of category practiced, which was demonstrated by the non-statistical difference in sex hormones between sprint and endurance athletes. Importantly, these athletes over 40 have hormonal levels similar to young adults of 34 years of age (Oddens & Vermeulen, 1996; Vermeulen & Oddens, 1996). The maintenance of these hormones in balance becomes an increasingly difficult task with advancing age (Kaufman & Vermeulen, 1997). Thus, the higher levels of these hormones in master athletes demonstrate a possible path to healthy aging and more years in sports participation at a highly competitive level (Storer et al., 2017).

Master athletes usually have a healthy diet, good stress management, and regular high-intensity exercise over many years, leading to several physiological adaptations responsible for hormonal production and maintenance (Korhonen et al., 2014). Appropriate body composition is a critical factor for athletic performance regardless of the type of event. Previous research shows that master athletes have higher lean mass and lower body fat than non-athlete controls (Simoes et al., 2017). Although MS had a higher level of lean mass and lower body fat, hormone levels were not different from ME. However, both groups fell within optimal body composition levels, which is reflected by all hormonal levels falling within reference values for younger adults.

Master athletes maintain greater lean mass and lower body fat (Swift et al., 2014), possibly due to T contributing to higher protein synthesis, leading to an increased resting metabolic rate and improved clinical markers such as lipid profile (Livingston et al., 2017). It was suggested that greater body fat, commonly observed in sedentary people over years, increases aromatase enzyme activity, converting T into ES (Xu et al., 2018), consequently reducing T and raising ES levels. Conversely, T triggers negative feedback to the hypothalamus for LH secretion (Börsch et al., 1975). Thus, the lower body fat observed in master athletes may be related to a reduction in T conversion to ES by the aromatase enzyme (Halbe, 1965; Lee et al., 2013) maintaining elevated T levels.

LH was 14.5% higher in T1 athletes. However, both groups were below reference values (T3: 16.0%; T1: 1.8%). Despite the large individual hormonal variation, this result may be explained due to a high T level, signaling the hypothalamus to decrease LH production and keep T in balance.

The high testosterone levels in the present sample, even balancing the LH through a negative feedback mechanism, suggests another via increasing T, as proposed by others (Lu et al., 1997). In the animal models, increased T in male rats during exercise might come from a direct stimulatory mechanism (independent of LH). In this case, lactate influences T secretion by increasing testicular adenosine production of 3', 5'-cyclic monophosphate (cAMP). This may partly explain why athletes experience a reduction in LH while maintaining high T. These mechanisms to maintain high T promote greater bone mineral density and libido, improved cognition, and may even contribute to the control of anxiety and depressive symptoms (Ari et al., 2004; Joshi et al., 2006; Maggio et al., 2013; McHenry et al., 2014; Tissandier et al., 2001; Walther et al., 2019).

Our results showed that IGF-1 was 19% higher in MS compared to ME athletes, which is consistent with previous studies in elite athletes (Dall et al., 2001) young (Elio et al., 2008) female (Eklund et al., 2021). Other investigators (Ehrnborg et al., 2003) found increased IGF-I induced by the maximum stress test for athletes involved in strength/speed sports, such as rowing, cycling, weightlifting, tennis, football, swimming, and decathlon. In addition, short-term exercises, such as the 30-second Wingate test or short-term all-out sprint, can cause an increase in the circulating level of IGF-I (Stokes et al., 2005). These short-duration and high-intensity exercise characteristics are common in master athletes' training routines, mainly MS.

In our analysis, we stratified athletes based on low (T1: \leq 81.1%) and high (T3: \geq 89.9%) sports performance. We found that body fat was 17.9% lower in the T3 group, indicating that body composition is related to athletic performance (Anderson, 1996). Better body composition (i.e., lower fat mass and higher lean mass) is a key factor in producing higher mechanical power output during each running stride. For example, less body mass leads to lower energy expenditure and attenuated fatigue (Anderson, 1996; Saunders et al., 2004).

On the other hand, the IGF-1 level of T3 athletes was 18.8% higher than T1 athletes. This increased IGF-1 level for the higher performance group may be due to intensified strength/power training sessions and higher-intensity training (Ehrnborg et al., 2003; Stokes et al., 2005). This difference may help to explain the increased lean mass observed for those with a higher level of IGF-1, such as MS in the present study. Greater lean muscle mass is known to benefit both speed (Benz et al., 2016) and long-distance (Rønnestad & Mujika, 2014) athletes, which corroborates the present results according to the positive correlation between performance and IGF-1.

Some limitations should be considered when interpreting the results of this study. First, due to the cross-sectional design, causation and temporal associations cannot be assumed. In addition, a larger sample size could provide more robust results. However, it is essential to note that we studied high-level master athletes, a particular small portion of the population, who are not widely available for scientific study. We did not assess objective measures of physical fitness (e.g., aerobic capacity, power, and/or strength) in our participants to determine how they relate to the outcome variables of interest. Due to the small sample size, we combined all participants in the secondary analyses. Thus, those results may be confounded given the higher levels of lean body mass and IGF-1 in the MS subjects. Finally, we do not know how these findings compare to master athletes in other activities, such as strength training events like powerlifting or bodybuilding.

Conclusion

In conclusion, MS athletes have a lower percentage of body fat and a higher IGF-1 level compared to ME athletes, which would suggest that strength/power exercise is more beneficial for these parameters in those groups of master athletes. In addition, higher IGF-1 was associated with better athletic performance for master athletes. The hormones LH, T, ES, and SHBG were similar between the two groups of athletes and were comparable to the reference values of younger adults. Thus, decades-long training can be a safe way to mitigate the effects of aging on the leading male hormones that are related to healthy functional aging. Further research would be useful to better understand the mechanisms by which a lifelong physical training, such as that of master athletes, can result in hormonal balance and maintenance of a healthy body composition.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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