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ORIGINAL RESEARCH

Acute Weight Training-Induced Testosterone Responses of Trained Males across Age Groups and Diets: A Pilot Study

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Abstract

Testosterone has been associated with health and athletic performance. However, it is also known to decrease with age. The rise of these age-related, non-communicable diseases affects economic growth. To develop natural, safe, and sustainable fitness and nutrition programs to keep the aging population healthy and fully functional, the present study explored exercise, diet, and age as possible factors affecting hormonal levels and responses. Twelve recreationally trained men from different age groups (20s to 70s) and diets (vegans and meat eaters) completed a 30-minute weight training protocol. Blood samples were taken before and after exercise to determine exercise-induced changes in total testosterone (TT) levels. Additional hormonal tests for cortisol (C) and testosterone-cortisol ratio (T/C) were conducted for outliers to guide future research. Pretest-posttest analysis showed a statistically significant increase in TT within subjects; $t(11)=-3.842$, $p=0.003$. Younger men (35 years old and below) had a significantly greater increase in TT compared to older men (40 years old and above); $X^2(1)=4.121$, $p=0.042$. Age was negatively correlated with TT increase ($r=-0.622$, $p=0.031$). Vegans showed higher estimated marginal mean TT levels in both pretest and posttest. In conclusion, a single session of 30-minute, moderate intensity, high-volume leg exercise can significantly increase blood serum TT in men across age groups and diets. Younger men tend to show greater increases in TT compared to older men. Further studies are needed to explore veganism as a more favorable diet for optimal testosterone levels.

Keyword: Acute testosterone response, weight training and testosterone, vegan diet and testosterone.

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INTRODUCTION

Background of the Study

Testosterone has been associated with muscle growth, strength, and various health measures, such as body composition (Rodriguez-Tolr, Torremadé, del Rio, di Gregorio & Miranda, 2013), bone density (Bouloux et al., 2013), brain function (Kocoglu et al., 2011), and immunity (Gold, Chalifoux, Giesser & Voskuhl, 2008). As a sex hormone, it is also linked with sexual drive and function (Gades et al., 2008). Recent studies on athletes also support the belief that testosterone plays an important role in athletic performance (Majumdar & Srividhya, 2010), recovery (Kargarfard, Amiri, Shaw, Shariat, & Shaw, 2018), fatigue (Bosco, Colli, Bonomi, Von Duvillard & Viru, 2000) and motivation (Crewther, Carruthers, Kilduff, Sanctuary & Cook, 2016). When it comes to fatigue and recovery, a low testosterone-cortisol ratio (T/C) is usually indicative of overtraining and catabolic status, while a high T/C marks anabolic status.

Unfortunately, testosterone levels in the body are known to decrease with age. Total testosterone (TT) levels fall at an average of 1.6% per year, while free and bioavailable levels fall by 2–3% per year among men (Feldman et al., 2002). Across age groups, clinically low testosterone levels (hypogonadism) was approximately 20% over age 60, 30% over age 70, and 50% over age 80 (Harman, Metter, Tobin, Pearson & Blackman, 2001). Furthermore, age-related hypogonadism is associated with erectile dysfunction, Type 2 diabetes, obesity, metabolic syndrome, osteoporosis, HIV, depression, cerebrovascular, and cardiovascular disease (Lunenfeld, Arver, Moncada, Rees & Schulte, 2012).

The World Health Organization estimates that around 1.5 billion of the world's population will be 65 years of age or older by 2050. In developing countries like the Philippines, the rise of these chronic, age-related, non-communicable diseases greatly affects economic growth (World Health Organization, 2011). Given the role of testosterone in health and the increase in older population, it is imperative to find safe and sustainable solutions to manage the natural decline of hormonal levels in aging men.

Review of Related Literature

Exercise Programming for Testosterone

There is a popular belief that resistance training can maintain health and virility by slowing down age-related decline in testosterone levels. Most studies on exercise programming for testosterone, however, involve young men.

Training volume. For instance, a study on young male recreational weightlifters found that a high-volume resistance training protocol for hypertrophy (10 sets of 10 repetitions, 75% 1RM, 2-minute rest periods, controlled movements) elicited significant elevations in salivary testosterone levels after a single session of exercise (Crewther, Cronin, Keogh & Cook, 2008). However, a study on college-aged men and women found that even a low-volume (2 sets, 90-second rest period) but high-speed resistance training protocol performed at maximal effort elicited significant increases in salivary testosterone after a single session of exercise (Caruso et al., 2012).

Rest period. In terms of rest period, a study on young male recreational weightlifters found that serum testosterone concentrations were significantly higher post-exercise in protocols with longer rest periods between sets (120 and 90 seconds) compared to one with a shorter rest (60 seconds) after a single session of exercise (Rahimi, Qaderi, Faraji & Boroujerdi, 2010).

Speed of contraction. Another factor that greatly affects exercise-induced testosterone levels is the speed of muscle contraction. A study on young male recreational weightlifters found that lifting weights at maximal and submaximal movement velocities (70% of maximum velocity) both significantly increased serum testosterone levels after a single session of exercise, given equal training volumes (Smilios, Tsoukos, Zafeiridis, Spassis & Tokmakidis, 2014). This was supported by another study on young male competitive strength and powerlifters, which reported significant elevations in post-exercise salivary testosterone during the power training phases of different periodization programs (Bartolomei et al., 2016).

Muscle group. But perhaps the most important consideration in programing for higher testosterone levels is the muscle group involved. A hypertrophy protocol involving upper body exercises alone did not result in significant elevations in both total and free testosterone levels (Migiano et al., 2010; Simão et al., 2013). In contrast, doing leg exercises alone (squat and leg press) resulted in significant post-exercise elevations in salivary testosterone in trained men (Crewther, Cronin, Keogh & Cook, 2008).

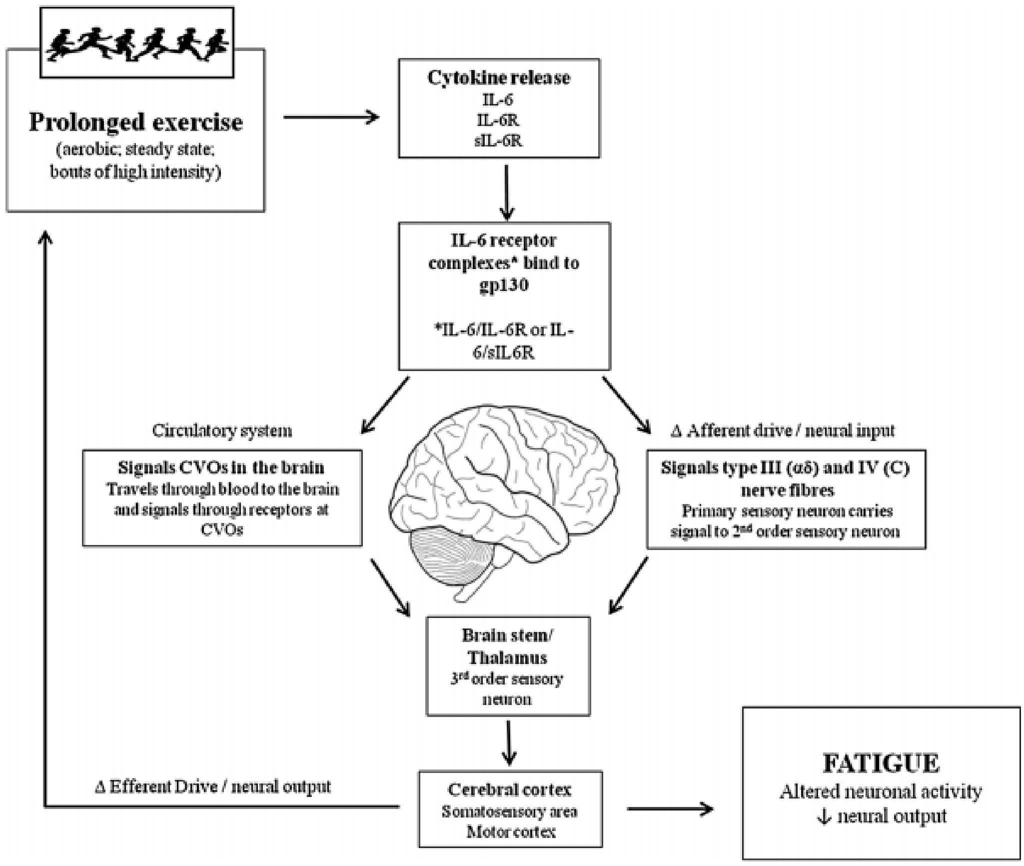


Figure 1. The Neuroinflammatory Model of fatigue in sport and exercise performance (Vargas & Marino, 2014).

Diet for Testosterone

Neuroinflammatory Model of fatigue. The negative effect of inflammation on exercise and sport performance has been well-studied (Kreher & Schwartz, 2012; Pyne, Hopkins, Batterham, Gleeson & Fricker, 2005; Robson-Ansley, Blannin & Gleeson, 2007). A review on fatigue mechanisms discussed the role of the inflammatory system in sport and exercise performance (Vargas & Marino, 2014). In the Neuroinflammatory Model, fatigue during exercise can be likened to fatigue during disease, which is characterized by the release of inflammatory markers like cytokines. Specifically, increase in IL-6 and other inflammatory mediators may decrease efferent drive to the muscles, resulting to impaired performance. The same review concluded that exercise may help improve the overall inflammatory profile of people with diseases, as well as blunt the inflammatory response of healthy individuals.

Using the Neuroinflammatory Model of fatigue, a diet that reduces inflammation may lead to better sport and exercise performance, which can induce higher testosterone levels.

Indeed, vegan men have been found to have higher TT levels than that of vegetarians and meat eaters (Allen, Appleby, Davey & Key, 2000). The same study found that compared to vegetarians and meat eaters, vegan men had significantly lower levels of IGF-1 hormone, which has been linked to prostate cancer.

The largest study on vegan diet to date, known as "Adventist Health Study 2", also concluded that a vegan diet that excludes meat and other animal products conferred lower risk for all kinds of cancers studied, when compared to other dietary patterns (Tantamango-Bartley, Jaceldo-Siegl, Fan & Fraser, 2013). This supported an earlier meta-analysis which concluded that compared to meat eaters, non-meat eaters had a significantly lower incidence of all kinds of cancers studied (Huang, Yang, Zheng, Li, Wahlqvist & Li, 2012).

Meanwhile, the latest long-term study on older adults (aged 45-74) found that higher intake of plant protein was associated with lower total mortality (Budhathoki, Sawada & Iwasaki, 2019). The same study found that substituting animal protein with plant protein was associated with lower risk of death from cancer and heart disease.

There are three possible mechanisms by which a vegan diet may help improve health and athletic performance: 1) less inflammation due to higher intake of antioxidants; 2) less inflammation due to less intake of inflammatory compounds; and 3) longer length of chromosomal telomere.

Higher intake of antioxidants. It is now well-established that intense and prolonged exercise can result in oxidative damage to both proteins and lipids in contracting muscle cells (Powers & Jackson, 2008). Hence, a diet rich in antioxidants can be beneficial in sport and exercise performance.

Compared to an omnivore diet, a vegan diet was associated with higher levels of antioxidants and omega 3 that are known to decrease inflammation, which has been linked to a range of diseases, including cancer (Miles et al., 2019). Another study reported an inverse relationship between prostate cancer risk and dietary intake of flavonoids, which have antioxidant effects, from fruits and vegetables (Russo et al., 2018).

In an experimental study on female swimmers, supplementation with phytoestorgens, a compound found in plants, enhanced antioxidant enzymes after exercise, as well as modulated sex hormone plasma levels (Mestre-Alfaro et al., 2011). Another experimental study on male runners found that supplementation with an antioxidant drink containing vitamin C and E alleviated exercise-induced oxidative damage in lymphocytes, but without blocking the cellular adaptation to exercise. (Sureda et al., 2008).

Less intake of inflammatory compounds. Processed meat intake was inversely associated with telomere length, which reflects oxidative and inflammatory responses (Nettleton, Diez-Roux, Jenny, Fitzpatrick & Jacobs, 2008). The high heat-generated glycotoxins in animal products, including meat, egg yolk, dairy, and their derivatives, increase cell-oxidant stress and promote inflammation (Vlassara, Cai & Crandall, 2002). Based on studies like these, the World Health Organization has classified red meat and processed meat as carcinogenic (World Health Organization, 2015).

Longer telomere length. Telomeres, the protective ends of chromosomes, shorten with age and disease. Oxidative stress accelerates telomere shortening, while antioxidants decelerate it (Kurz, Decary, Hong, Trivier, Akhmedov & Erusalimsky, 2004; Von Zglinicki, 2002). Indeed, an experimental study found that age-dependent telomere shortening can be decelerated by suppressing oxidative stress through intake of Vitamin C (Furumoto, Inoue, Nagao, Hiyama, & Miwa, 1998), a micronutrient that has been found to be higher among vegans compared to the general population (Kristensen et al., 2015). In another study, consumption of plant-based foods like seeds, nuts, legumes, seaweeds, and coffee was associated with longer telomeres (Freitas-Simoes, Ros & Sala-Vila, 2016).

It is the goal of this study to compare testosterone levels of male vegans to that of meat eaters as an acute response to weight training. Also, men of different age groups (20s, 30s, 40s, 50s, 60s, and 70s) were included to explore age-related trends in exercise-induced hormonal responses.

Theoretical Framework

Based on the aforementioned studies on exercise, nutrition, and aging, the author formulated the following theoretical models.

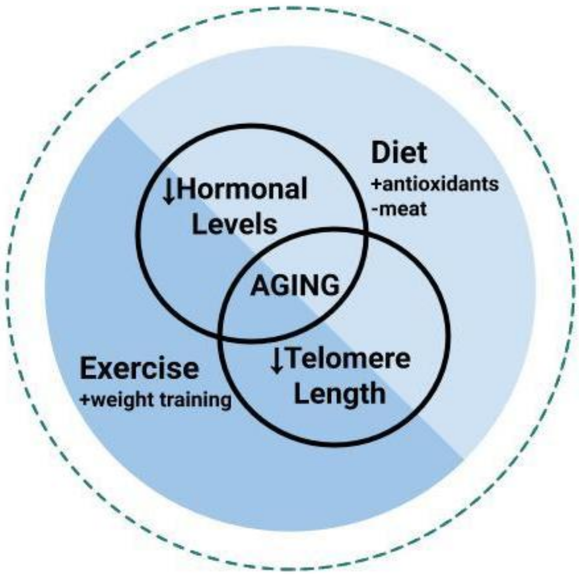


Figure 2. The author's general theoretical framework showing the two known measures of aging (telomere length and hormonal levels) and the two known lifestyle factors affecting them (diet and exercise). For diet, antioxidant intake is a favorable factor in slowing down aging, while meat intake is an aggravating factor for aging. For exercise, weight training is a favorable factor in slowing down aging.

METHOD

Design

Participants were randomly assigned to a training schedule between 11AM to 2PM. Lunchtime was chosen since cortisol levels have been found to peak in the morning, while testosterone levels do not vary significantly throughout the day among apparently healthy males (Hayes, Grace, Kilgore, Young & Baker, 2012). Vegans and non-vegans were tested together to minimize experimenter bias.

All participants followed a moderate-intensity hypertrophy protocol previously found to elicit significant elevations in testosterone levels: 10 total sets of 10RM, 75% 1RM, 2 minutes rest between sets (Crewther, Cronin, Keogh & Cook, 2008). A previous study on male seniors aged 65-70 also employed the said training protocol (Häkkinen, Kraemer, Pakarinen, Triplett-McBride, McBride & Häkkinen, 2002). Such moderate-intensity protocol was chosen over the high-speed, high-intensity training of Caruso et al. (2012) to ensure safety of older participants. Also, a study found that high-intensity training led to decreased testosterone levels (Abdollahzadeh & Ashrafizadeh, 2018).

One week before testing day, assessment of 10RM loads for each participant was done following guidelines of the American Council on Exercise (Jimenez, 2018). Participants were also requested to avoid cardiovascular endurance training and vigorous physical activity for 48 h, abstain from consuming substances known to affect performance (alcohol, caffeine, creatine, whey protein, pre-workout, etc.) for 24 h, and get quality sleep for at least 6 h the night before testing. They were also instructed to maintain their regular diet throughout the days leading to the study.

On testing day, blood samples were taken right before and right after exercise by the same medical technologist to minimize procedural variability. Participants were also asked to rate the difficulty of the exercise using the Borg Ratings of Perceived Exertion Scale.

Subjects

A total of 12 men consisting of six vegans and six meat eaters (mean age=39.92±15.05; mean BMI=24.51±0.92) who practiced resistance training regularly for recreational purposes during the time of study were included.

Inclusion Criteria

Inclusion criteria involved factors that are known to affect TT levels, such as: being apparently healthy, of normal weight, non-smoker, not on any medication or supplementation known to affect testosterone levels (testosterone boosters, Viagra, etc.), not on any supplementation known to affect performance (creatine, preworkout, etc.), never used anabolic steroids, not currently training for cardiovascular endurance, not currently active in any sport, not chronically sleep deprived, not chronically stressed, and not working in the night shift.

For vegan subjects, a minimum of one year of practicing a vegan diet was required to participate in the study. Only those who were vegans for ethical reasons (e.g. vegan for the animals, vegan for religion, etc.) for at least one year were included to ensure that they did not "cheat" on their vegan diet.

Sampling Method

Participants were recruited from different gyms and from online vegan and fitness groups where the researcher posted advertisements. They were asked to answer an online survey to determine if they were fit for the study. An online interview was also conducted by the author to supplement the said survey.

Ethical Considerations

Each subject signed a standard Physical Activity Readiness Questionnaire form, as well as an institutionally approved informed consent form to participate in the study. The current study was approved in advance by a panel of professors in the University of the Philippines College of Human Kinetics Graduate Studies Department.

Locale of the Study

The pilot study was done in an air-conditioned fitness gym in Quezon City, Philippines, from April 22-27, 2019. Room temperature was set at 20 degrees Celsius, while humidity was set at 42%, which were well within ACSM recommendations for exercise (Armstrong, Casa, Millard-Stafford, Morán, Pyne & Roberts, 2007; No & Kwak, 2016).

Instruments of the Study

1. Life Fitness Hammer Strength 45 Degree Linear Leg Press with Weights - exercise machine used for the leg press

2. Hammer Strength HD Athletic Power Rack with Standard Olympic Bar and Weights - exercise equipment used for the barbell back squat

3. iPhone 6s - used as timer and alarm for participants

Exercise Protocol

The protocol was the same for all subjects: Two compound leg exercises (Smith machine barbell back squat and supine leg press), each performed for five sets of 10RM (75% 1RM) with 2-minute rest periods between sets, and in controlled movements with 1.5 seconds concentric and 1.5 seconds eccentric movement (Crewther et al., 2008). The entire protocol, including warmup, lasted for approximately 30 minutes.

When participants felt fatigued and were unable to maintain 75% 1RM for 10 repetitions, the load was reduced to ensure five sets of 10 repetitions could still be completed.

Before starting the first set of exercises, subjects performed a standard warm-up consisting of five minutes of leg mobility drills, and two submaximal sets on the leg press machine (20 repetitions, 50% 1RM).

Participants were tested at the same air-conditioned gym with constant temperature (20 degrees Celsius) and humidity (42%) at the same time of day near lunchtime (11AM to 2PM) to avoid extreme hormonal levels that may be caused by diurnal variations. To minimize procedural variability, the same investigator/coach supervised all testing sessions and provided verbal encouragement during exercise.

Blood Tests

Blood samples were collected from the left antecubital fossa of the participants while they were seated before warm-up, and right after the last repetition of the last exercise. To minimize procedural variability, the same medical technologist collected blood samples from the antecubital fossa of all participants. Whole blood was centrifuged (Hsiangtai, Taiwan) for 10 minutes, after which serum was aspirated, aliquoted, and analyzed for TT immediately (Cobas E 411, Roche Diagnostics, USA). Blood samples were stored in the laboratory for follow-up tests. One week after testing, additional hormonal tests for cortisol (C) and T/C were requested to explain the performance of outliers.

Subjects were instructed to have their normal, complete meals 2 h before testing. During the exercise protocol, they were allowed to drink water as needed.

Statistical Analyses

Data are presented in means ± standard deviation, and significance level was set at alpha 0.05. Prior to statistical analysis, dependent variables were tested for normality using Shapiro-Wilk test, and for homogeneity using Levene's test. Data showed normal distribution, but the assumption of homogeneity was violated for TT increase. Hence, non-parametric Kruskal-Wallis H was used to analyze between-group differences, while paired samples t-test was used to analyze within-subject differences in TT scores. Pearson correlation was used to explore relationships between variables.

RESULTS

Within Subjects

All subjects had normal baseline levels of TT pretest. Pretest (M=494.44, SD=177.27) and posttest (M=560.19, SD=203.05) statistical analysis showed a significant increase in TT scores within subjects; $t(11)=-3.842$, $p=0.003$. Participants rated the exercise protocol as "hard" (mean RPE=15.42±2.11).

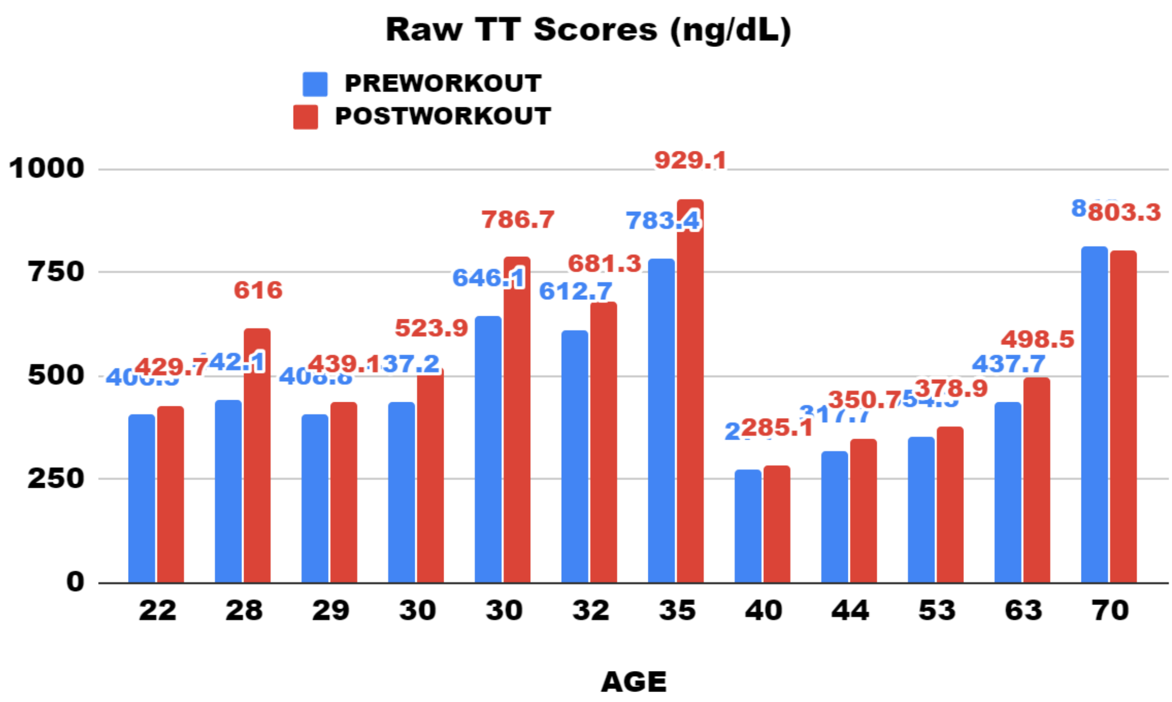


Figure 3. Raw TT scores of participants (N=12) according to age, with a statistically significant increase within subjects; $t(11)=-3.842$, $p=0.003$.

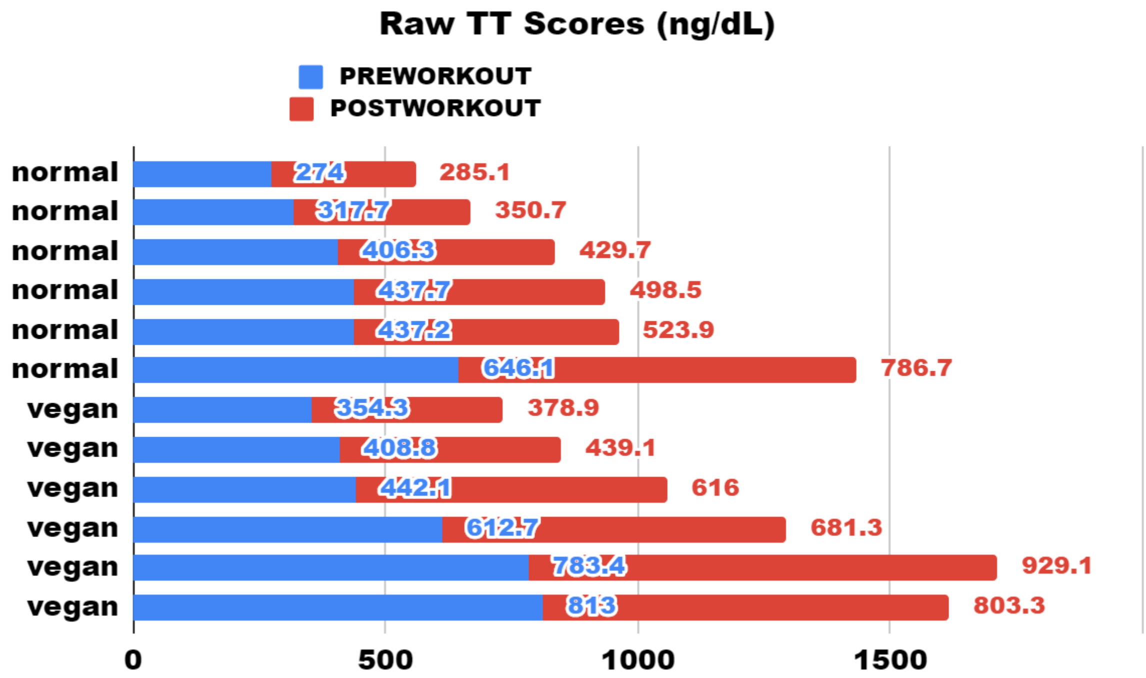
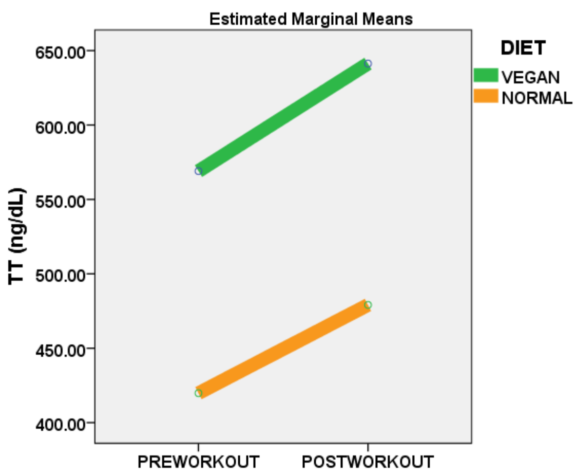


Figure 4. Raw TT scores of participants (N=12) according to diet.

Between Groups

Between-groups statistical analysis showed younger men (35 years old and below) had a significantly greater increase in TT scores compared to older men (40 years old and above); $X^2(1)=4.121$, $p=0.042$, with a mean rank pretest-posttest difference of 8.29 (N=7) for younger men and 4 (N=5) for older men. Age group was negatively correlated with TT increase ($r=-0.622$, $p=0.031$).

Vegans had higher estimated marginal mean TT levels in both pretest and posttest. Diet was almost correlated with percentage increase in TT ($p=0.054$).



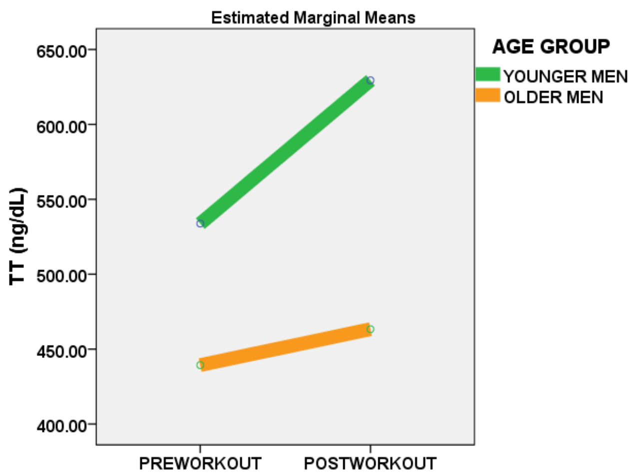


Figure 5. Estimated marginal means of TT responses of younger men (N=7) versus older men (N=5) with a statistically significant difference ($p<0.05$) between groups (left). Estimated marginal means of TT responses of vegans (N=6) versus meat eaters (N=6) with no statistically significant difference between groups (right)

Outliers

The outlier in the younger men's group (age=22) who had the least increase in TT (TTPRE=406.3 ng/dL, TTPOST=429.7 ng/dL) also exhibited an increase in C (CPRE=14900 ng/dL, CPOST=18470 ng/dL) and a decrease in T/C (T/CPRE=0.03 ng/dL, T/CPOST=0.02 ng/dL).

Meanwhile, the outlier in the older men's group (age=70) showed a decrease in both TT (TTPRE=813 ng/dL, TTPOST=803.3 ng/dL) and C (CPRE=71500 ng/dL, CPOST=58700 ng/dL), and an increase in T/C (T/CPRE=.11 ng/dL, T/CPOST=.14 ng/dL).

CONCLUSION AND DISCUSSION

The aim of this pilot study was to explore how men of different age groups and diets respond to a 30-minute, moderate-intensity weight training session designed to elicit maximal TT. To the author's knowledge, this is the first of its kind to compare meat eaters and vegans in terms of acute testosterone response to weight training.

It was observed that 11 out of 12 participants showed an increase in TT following weight training, and this was statistically significant within subjects; $t(11)=-3.842$, $p=0.003$. This supports several studies that found a single bout of high-volume hypertrophy scheme to be effective in increasing testosterone levels (Linnamo, Pakarinen, Komi, Kraemer & Hakkinen, 2005; Crewther et al., 2008).

To make sense of the performance of outliers, follow-up hormonal tests for C and T/C were done. Being the "stress hormone", C has long been used as an indicator of fatigue and catabolic nature of exercise. Hence, higher C would lead to a lower T/C which has been correlated with overtraining (Kargarfard, Amiri, Shaw, Shariat, & Shaw, 2018). Conversely, a higher T/C ratio indicates anabolic status and has been correlated with athletic performance (Majumdar & Srividhya, 2010).

Interestingly, the outlier in the young men's group (age=22) who exhibited the least TT increase also lifted the heaviest total weight and rated the protocol as "extremely hard" (RPE=19). The subject, a fitness coach and recreational powerlifter, reported a weight training experience of 10 years, and a lifting frequency of five times a week. Indeed, follow-up tests revealed that he had a corresponding increase in C and a decrease in T/C postworkout. It can be inferred that the minimal change in TT was mediated by fatigue.

A similar study on exercise-induced hormonal responses mentioned that across different protocols, total exercise stress may induce increases in C (Smilios, Tsoukos, Zafeiridis, Spassis & Tokmakidis, 2014). This provides an interesting take on the popular opinion of pushing one's limits during exercise. When the goal is increase in testosterone levels, it may help to keep the exercise intensity at moderate levels.

Meanwhile, the outlier in the older men's group and the oldest participant (age=70) who did not show an increase in TT was found to have a corresponding increase in both C and T/C. As his T/C ratio still increased, fatigue is less likely to be a cause of the decrease in TT. However, he rated the protocol as "very hard" (RPE=18). The subject, a retired senior, reported a weight training experience of one year and a lifting frequency of three times a week. However, he admitted that he rarely performs leg exercises.

In a similar study among older men (60-75 years old), acute increases in serum TT were not observed following the same resistance training protocol (Smith machine squat, 5 x

10RM, 2 minutes rest between sets) before the participants underwent a 24-week strength training program. However, after the 24-week training program, the same participants showed significant acute increases in TT (Häkkinen et al., 2002). It seems that training status mediates acute hormonal responses to exercise.

When it comes to diet, vegan men had consistently higher estimated marginal mean TT scores in both pretest and posttest compared to meat eaters. Diet was only almost significantly correlated with percentage increase in TT (p=0.054). A previous study found that a vegan diet was associated with higher TT levels compared to vegetarian and omnivore diets (Allen et al., 2000). Perhaps a significant result would have been achieved with a larger sample size.

In conclusion, a single session of 30-minute, moderate intensity, high-volume leg exercise can significantly increase blood serum TT across age groups and diets. Younger men tend to show greater increases in TT compared to older men. Further studies are needed to explore veganism as a more favorable diet for optimal testosterone levels.

Limitations

The current paper is a pilot study aimed at exploring various factors that can guide future studies. Hence, sample size was limited, and tests for C and T/C levels were done for the outliers only. Also, there was no control group, making the study more descriptive than experimental.

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