2021

Neutral Effect of Exenatide on Serum Testosterone in Men with Type 2 Diabetes Mellitus: A Prospective Cohort

Sky Graybill
US Army Brooke Army Medical Center

Jennifer Hatfield
US Army Brooke Army Medical Center

Maria Kravchenko
US Army Brooke Army Medical Center

Darrick Beckman
Wilford Hall Ambulatory Surgical Center

Joshua Tate
Keesler Air Force Base

See next page for additional authors

Received: 9 June 2020 | Revised: 21 December 2020 | Accepted: 30 December 2020

DOI: 10.1111/andr.12966

ORIGINAL ARTICLE

ANDROLOGY WILEY

Neutral effect of exenatide on serum testosterone in men with type 2 diabetes mellitus: A prospective cohort

Sky Graybill[1] | Jennifer Hatfield[1] | Maria Kravchenko[1] | Darrick Beckman[2] | Joshua Tate[3] | Alexis Beauvais[4] | Philip Clerc[1] | Desarae Davila[2] | Whitney Forbes[2] | Jana Wardian[2] | Matthew Kemm[1] | Abegail Hubberd[2] | Mark True[1]

[1]Medicine, US Army Brooke Army Medical Center, San Antonio, TX, USA

[2]Medicine, Wilford Hall Ambulatory Surgical Center, San Antonio, TX, USA

[3]Medicine, Keesler Air Force Base, Biloxi, MS, USA

[4]International Health Specialist HQ USSOUTHCOM, Command Surgeon's Office, Miami, FL, USA

Correspondence
Sky Graybill, MD (Brooke Army Medical Center; 3551 Roger Brooke Dr., Fort Sam Houston, TX 78234, USA.
Email: sky.d.graybill.mil@mail.mil

Funding information
The 59th MDW Chief Scientist's Office/Science and Technology, Grant/Award Number: Funded data analysis for this research.

Abstract

Background: Endogenous testosterone increases with weight loss from diet, exercise, and bariatric surgery. However, little is known about testosterone levels after weight loss from medication.

Objectives: Uncover the effects of Glucagon-Like Peptide-1 receptor agonist (GLP-1 RA) therapy on serum testosterone.

Material and Methods: Prospective cohort study of men starting GLP-1 RA therapy for type 2 diabetes mellitus.

Results: 51 men lost 2.27 kg (p = 0.00162) and their HbA1c values improved by 0.7% (p = 0.000503) after 6 months of GLP-1 RA therapy. There was no significant change in testosterone for the group as a whole. However, in subgroup analyses, there was a significant difference in total testosterone change between men starting with baseline total testosterone <320 ng/dL (238.5 ± 56.5 ng/dL to 272.2 ± 82.3 ng/dL) compared to higher values (438 ± 98.2 ng/dL to 412 ± 141.2 ng/dL) (p = 0.0172);free testosterone increased if the baseline total testosterone was <320 ng/dL (55.2 ± 12.8 pg/mL to 57.2 ± 17.6 pg/mL) and decreased if >320 ng/dL (74.7 ± 16.3 pg/mL to 64.2 ± 17.7 pg/mL) (p = 0.00807). Additionally, there were significant differences in testosterone change between men with HbA1c improvements ≥1% (351.6 ± 123.9 ng/dL to 394.4 ± 136.5 ng/dL) compared to men with HbA1c changes <1% (331.8 ± 128.6 ng/dL to 316.1 ± 126.2 ng/dL) (p = 0.0413).

Conclusion: GLP-1 RA therapy improves weight and HbA1c without adverse effects on testosterone. Those starting with lower testosterone values or attaining greater improvement in HbA1c may see additional benefits.

KEYWORDS
glucagon-like peptide-1, hypogonadism, testosterone

1 | INTRODUCTION

Male hypogonadism has become a common diagnosis, and testosterone therapy is often prescribed as a first line option without full consideration of adverse effects and alternative strategies to treat underlying causes. Potential complications of testosterone therapy may include polycythemia,[1] worsening of obstructive sleep apnea,[2] and cardiovascular disease.[3] An increased risk of stroke has been reported in several studies, particularly within the first year of testosterone therapy (3; 4). Cardiovascular risks or benefits of testosterone therapy are not proven and large randomized controlled trials with this focus are needed.[5] Men with diabetes mellitus have an increased risk of cardiovascular disease and hypogonadism,[6] raising concern about the safety of testosterone therapy in this population. Weight loss from diet, exercise, and bariatric surgery consistently increases serum testosterone in men with functional hypogonadism,[7] and may represent a preferred initial strategy over exogenous testosterone in select patients.[6]

In addition to diabetes mellitus, obesity and metabolic syndrome are associated with functional hypogonadism.[8] Weight loss in patients with these disorders is a fundamental component in disease management, improving both disease progression and other quality-of-life factors, to include gonadal function. Many diabetes medications also affect weight with a predictable impact on testosterone levels. For example, pioglitazone promotes weight gain and appears to lower testosterone; 50 eugonadal men taking pioglitazone for 6 months gained an average of +1.9 kg and their testosterone levels significantly decreased.[9] In contrast, metformin facilitates weight loss and is associated with increased testosterone. There was a study of 35 eugonadal men with metabolic syndrome who took metformin for 4 months. They lost an average of −2.3 kg, serum total testosterone increased on average by +44 ng/dL, and free testosterone increased on average by +4.6 pg/mL.[8] Glucagon-like peptide-1 receptor agonists (GLP-1 RA) typically result in a −2 to −3 kg weight loss in people over 3 to 12 months.[10] However, there is little information about how GLP-1 RA therapy affects testosterone. One study[11] showed a decrease in diurnal variations of serum testosterone in men taking GLP-1 RA therapy over a 24-hour period; this trial did not have a sufficient duration to affect weight. There was a retrospective study of 30 men with obesity, diabetes, post-puberty hypogonadism, and erectile dysfunction who had been making lifestyle changes as well as taking testosterone and metformin for a year.[12] Of this group, 16 men added liraglutide for an additional year. Their total testosterone values increased from 466.1 ± 63.6 ng/dL to 481.7 ± 57.3 ng/dL but their free testosterone did not significantly change. Their weight decreased from 99 ± 7.6 kg to 93.7 ± 6.3 kg, and HbA1c values improved from 8.3 ± 0.3% to 7.3 ± 0.3%. They also reported improvement in erectile function. A more recent study[13] showed exenatide therapy for 3 months had beneficial effects on serum testosterone.

It was unclear if weight loss from GLP-1 RA therapy stimulates testosterone production or if decreases in testosterone diurnal variations significantly impact testosterone levels. The aim of our study was to uncover the 6-month biochemical effects of GLP-1 RA therapy on serum testosterone.

2 | MATERIAL AND METHODS

This was a prospective, observational cohort study of men ≥18 years of age starting GLP-1 RA therapy for type 2 diabetes mellitus (T2DM). The participants received medical care at Brooke Army Medical Center (BAMC) or Wilford Hall Ambulatory Surgical Center (WHASC). The BAMC Institutional Review Board approved this study, and participants provided written informed consent. Exclusion criteria included[1]: previously diagnosed organic disease of the pituitary or testicles[2]; current use of exogenous testosterone, opioids, or leuprolide[3]; GLP-1 RA therapy within the previous 12 months; and [4] a continued need for diabetes medication not indicated for use with GLP-1 RA therapy (dipeptidyl peptidase-4 inhibitors). Participants could be on other medications for diabetes. Although any GLP-1 RA was permitted in our study, exenatide extended-release (exenatide ER) was the formulary agent, so it was the predominant medication evaluated. The decision to include male patients as young as 18 years of age, as opposed to only older male patients with T2DM, was made with consideration of the rising rates of T2DM in the U.S. population. The 2020 National Diabetes Statistics Report highlighted the continued increased incidence rates of T2DM in children and young adults.[14] Obesity has become more prevalent in younger males over time as well. Males younger than 18 years of age were not included in this study, due to differences in normal testosterone ranges for pediatric patients.

Eligible participants met with a member of the study staff to obtain written informed consent and schedule fasting baseline laboratory studies. They provided Health Insurance Portability and Accountability Act (HIPAA) authorization to allow study staff to collect study-related data from the electronic medical record. Participants' names and identifiers were recorded in a master key associated with a unique identifier to connect data to the participants.

Each participant received at least 6 months of GLP-1 RA therapy to allow sufficient time for GLP-1 RA to reach full therapeutic effects and for patients to reach their nadir weight. Patients in the study were followed for up to 12 months. Study staff collected the following information during screening and after at least 6 months of GLP-1 RA therapy: height (baseline only), weight, HbA1c, total testosterone, calculated free testosterone, sex hormone binding globulin (SHBG), albumin, current medications, and other medical conditions. The calculation for free testosterone was the Vermeulen equation. Free testosterone = ([Testosterone] − (N × [FT]))/(Kt{SHBG − [Testosterone] + N[free T fraction]}). Kt is the association constant of SHBG for Testosterone. N = KaCa+1. Ka is the association constant of albumin for Testosterone, and Ca is the albumin concentration.

Participants' data were stored on a limited access, password-protected computer network.

Power analysis estimated means and standard deviations for total testosterone, free testosterone, and SHBG based on data in a prior

study about testosterone before and after 6 months of weight loss from lifestyle changes.[15] In that study, participants lost on average −16.3 kg. SHBG went from 27.6 ± 11.9 nmol/L to 32.6 ± 12.9 nmol/L; the pooled standard deviation was 12.4 nmol/L, and effect size was 0.403 so the power analysis was that the number of participants per group would need to be 51 paired. Free testosterone went from 185 ± 66 pmol/L to 212 ± 84 nmol/L; pooled standard deviation was 75 pmol/L, and effect size was 1.7 so the power analysis was that the number of participants per group would need to be 5 paired. Total testosterone went from 12 ± 4 nmol/L to 14 ± 4 nmol/L; the pooled standard deviation was 4 nmol/L, and effect size was 0.5 so the power analysis was that the number of participants per group would need to be 34 paired. SPSS Sample Power 3.0 (IBM) estimated the required sample size for a power of 80% with a level of confidence of 95%. A correlation coefficient of <−0.50 was considered a good index of an inverse relationship between body weight and serum testosterone. According to the method of Kraemer and Thiemann,[16] detecting a correlation coefficient of <−0.50 required 51 participants.

Primary outcome variables were total testosterone, SHBG, and calculated free testosterone. Paired t-tests compared these variables at baseline and after at least 6 months of GLP-1 RA therapy.

3 | RESULTS

Between 2015 and 2018, 79 participants enrolled; of these, 51 participants completed the study. Reasons for drop out included: Principal investigator withdrew 14 for protocol deviation (incomplete baseline laboratory testing prior to GLP-1 RA initiation), 8 were lost to follow-up, 4 withdrew consent, 1 had GLP-1 RA therapy stopped by therapeutic provider due to nausea and vomiting, and 1 died for reasons unrelated to the study or medication.

Table 1 shows participant baseline characterizes. All participants started exenatide extended-release (ER) 2 mg weekly due to formulary restrictions. One participant switched to dulaglutide 0.75 mg weekly after 3 months, and the remainder continued exenatide ER 2 mg weekly throughout the study.

Over the study period, participants on average lost −2.27 kg (p = 0.00162). BMI at baseline was 34.9 ± 5.3 kg/m^2 and at follow-up was 34.3 ± 5.7 kg/m^2. HbA1c values improved on average by 0.7% (p = 0.000503). For the group as a whole, there was no significant change in total testosterone; baseline values were 334 ± 126 ng/dL and follow-up values were 339 ± 133 ng/dL (p = 0.710). Similarly, free testosterone was not significantly different with baseline values of 64 ± 17 pg/mL and follow-up values of 60 ± 18 pg/mL (p = 0.221). However, SHBG significantly increased from 36 ± 15 nmol/L at baseline to 41 ± 22 nmol/L (p = 0.0285) (Table 2).

Subgroup analysis showed no significant difference in testosterone change between the groups that lost ≥3% of their total body weight (321.8 ± 117.6 ng/dl to 323.6 ± 120.6 ng/dl) compared to those that lost <3% (343.0 ± 131.4 ng/dl to 340.5 ± 141.5 ng/dl) (p = 0.244) (Figure 1).

TABLE 1 Baseline characteristics (n = 51).

Age in years, mean ±SD	57.6 ± 8.8
Race	
Caucasian/White, n (% of group)	38 (74.5%)
African American/Black, n (% of group)	9 (17.6%)
Native American, n (% of group)	2 (3.9%)
Other, n (% of group)	2 (3.9%)
Ethnicity	
Hispanic, n (% of group)	15 (29.4%)
Non-Hispanic, n (% of group)	36 (70.6%)
Diabetes duration, mean ±SD (years)	9.8 ± 7.3
Diabetes medications	
Metformin, n (% of group)	46 (92%)
Insulin, n (% of group)	42 (82%)
Sodium–glucose co-transporter 2 inhibitor, n (% of group)	7 (14%)
Thiazolidinedione, n (% of group)	2 (4%)
Sulfonylurea, n (% of group)	5 (10%)
Dipeptidyl peptidase IV inhibitor stopped upon enrollment, n (% of group)	42 (82%)
Obstructive sleep apnea, n (% of group)	13 (25%)
Erectile dysfunction, n (% of group)	14 (27%)

Abbreviation: SD, standard deviation.

TABLE 2 Measurements at baseline and 6 months (n = 51).

	Baseline mean ±SD	6 Months mean ±SD	p-value
Body weight (kg)[b]	110 ± 20	108 ± 21	0.00162
BMI (kg/m^2)[b]	34.9 ± 5.3	34.3 ± 5.7	0.00161
[a]HbA1c (%)[b]	8.1 ± 1.5	7.4 ± 1.4	0.000503
Total testosterone (ng/dL)	334 ± 126	339 ± 133	0.710
SHBG (nmol/L)	36 ± 15	41 ± 22	0.0285
Free testosterone (pg/mL)	64 ± 17	60 ± 18	0.221

Abbreviations: BMI, body mass index; SD, standard deviationSHBG, sex hormone binding globulin.

[a][10.93 × A1c%]−23.5 = mmol/mol.

[b]Notifies significant change.

Subgroup analysis did show a significant increase in total testosterone for men who had an HbA1c improvement ≥1% compared to those with HbA1c change <1% (p = 0.0413). HbA1c improvement of ≥1% occurred in men with higher baseline HbA1c (mean value of 8.96% versus 7.7%). Total testosterone increased from 351.6 ± 123.9 ng/dL to 394.4 ± 136.5 ng/dL for men with HbA1c improvement ≥1%. In contrast, total testosterone decreased from the baseline of 331.8 ± 128.6 ng/dL to 316.1 ± 126.2 ng/dL for men with a HbA1c change <1% (Figure 1). There was no significant change in free testosterone from baseline regardless of A1c change (66.0 ± 22.0 pg/mL to 66.0 ± 23.8 pg/mL if ≥1% vs. 64.2 ± 14.7 pg/mL to 57.45 ± 13.4 if <1%) (Figure 2).

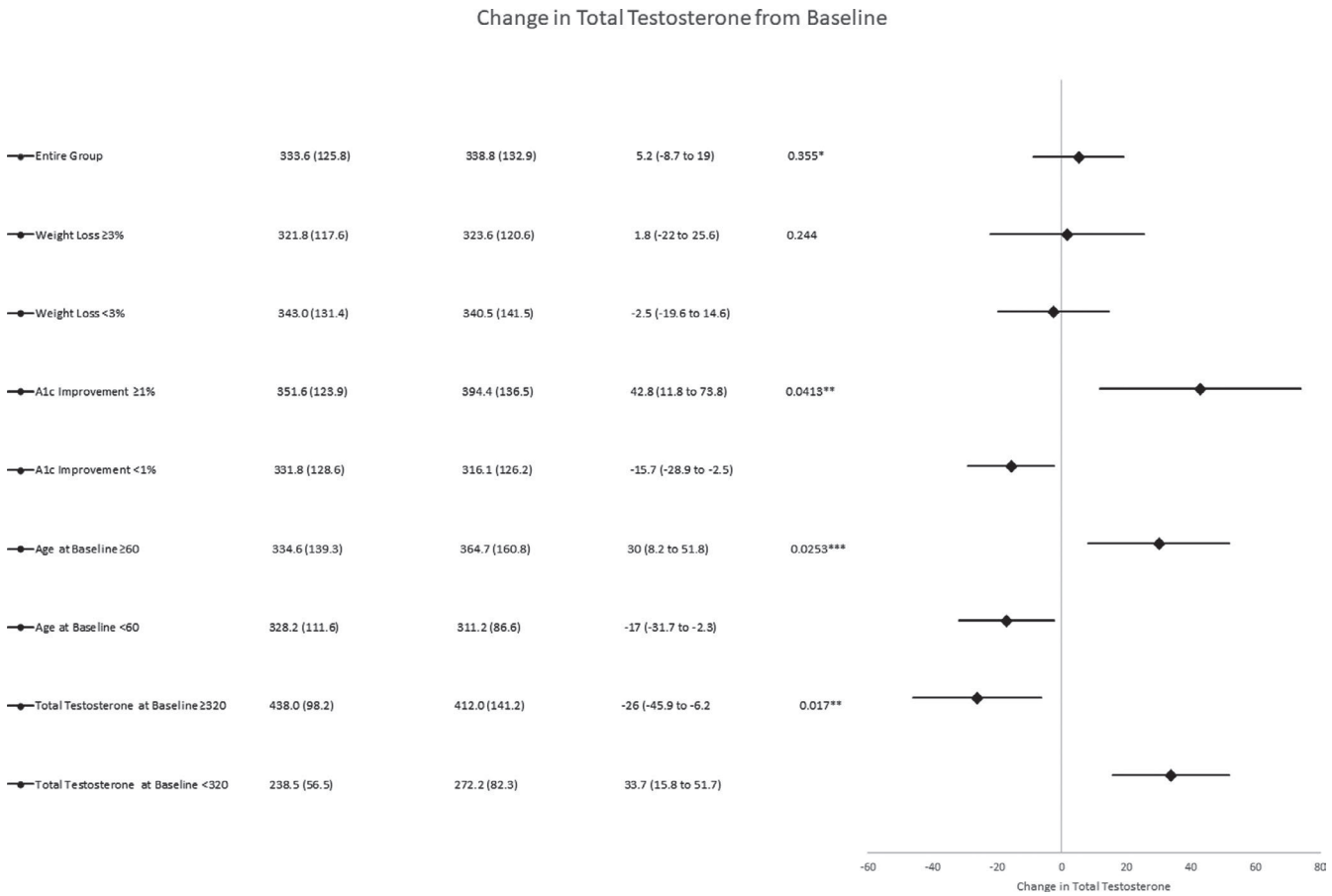

FIGURE 1 Change in Total Testosterone from Baseline. The horizontal bars represent standard error of the mean. The vertical line represents no change from baseline. P-values are one sided t-tests assessing the change in Total Testosterone between subgroups. *Two-sample, one-sided paired t-test assessing decrease between the baseline and 6-month values for the entire group. **Notifies significant change. ***Using the non-parametric Wilcox test resulted in p-value = 0.0578

Figure 3 depicts scatter plots of change in total and free testosterone by change in weight and HbA1c for the entire study group. There were large variations in testosterone values among participants. Notably, there was no significant relationship between testosterone levels and weight loss. The strongest, but not significant, relationship was between improvements in total testosterone and improvements in HbA1c.

Figure 4 depicts bar graphs comparing men who had HbA1c improvements ≥1% to those with HbA1c change <1%, breaking it down by change in total testosterone, free testosterone, SHBG, and weight. This analysis also showed that change in testosterone was not related to the small weight loss in this study. The bar graphs did show a significant change for total testosterone with HbA1c improvement ≥1% compared to HbA1c change <1%; it did not show this relationship for free testosterone. The change in SHBG accounted for the differences between total and free testosterone; the increase in SHBG correlated with the increase in total testosterone while the free testosterone did not change.

Initially, subgroup analysis by t-test appeared to show a difference for participants who were older or younger than 60 years of age; older participants appeared to have greater increases in testosterone. However, the confirmatory Wilcoxon test did not show statistical significance (Figure 1).

Interestingly, subgroup analysis showed a statistically significant difference for men whose baseline testosterone was <320 ng/dL compared to ≥320 ng/dl (p = 0.0172). We selected this threshold because one study of 3369 participants[17] showed that symptoms of sexual dysfunction were more likely to be present when total testosterone values were less than 320 ng/dL. In our study, total testosterone increased from 238.5 ± 56.5 ng/dL to 272.2 ± 82.3 ng/dL in men with baseline total testosterone values <320 ng/dL. Total testosterone started at 438 ± 98.2 ng/dL and decreased to 412 ± 141.2 ng/dL for men with higher baseline total testosterone values (Figure 1).

Similarly, the change in free testosterone was significantly different between men whose baseline testosterone was <320 ng/dL compared to ≥320 ng/dL (p = 0.00807). Free testosterone increased from 55.2 ± 12.8 pg/mL to 57.2 ± 17.6 pg/mL for men with baseline total testosterone values <320 ng/dL. Free testosterone started at 74.7 ± 16.3 pg/mL and decreased to 64.2 ± 17.7 pg/mL for men with higher baseline total testosterone values (Figure 2).

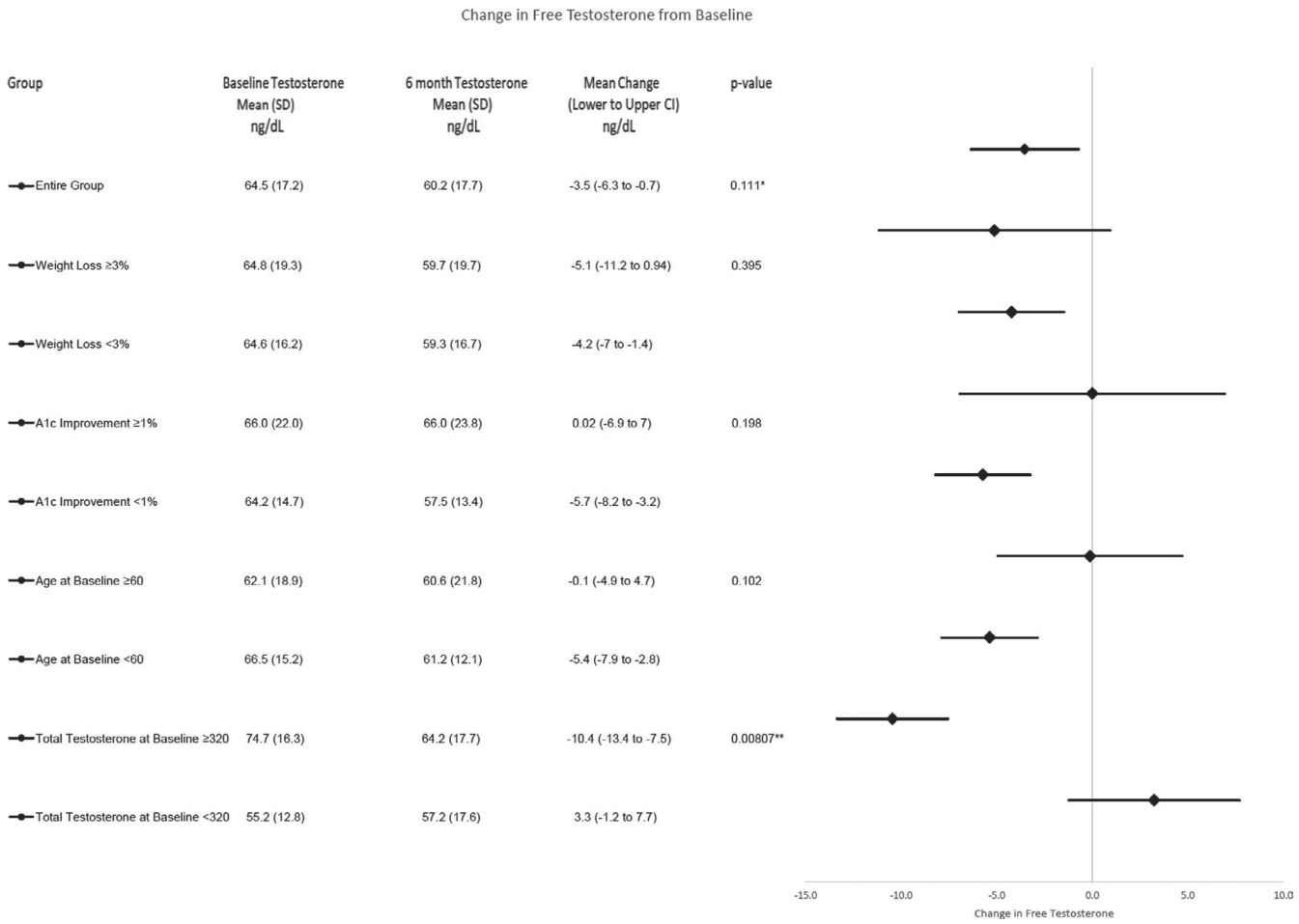

FIGURE 2 Change in Free Testosterone from Baseline. The horizontal bars represent standard error of the mean. The vertical line represents no change from baseline. P-values are one sided t-tests assessing change in Free Testosterone between subgroups. *Two-sample, one-sided paired t-test assessing increase between the baseline and 6-month values for the entire group. **Notifies significant change versus those with a total testosterone ≥320 mg/dl at baseline

4 | DISCUSSION

Our study mirrors the results of a trial where 18 healthy men received GLP-1 RA therapy over an 8-hour infusion and a placebo infusion on separate days.[18] Participants in this placebo-controlled crossover study ate less after GLP-1 RA therapy than after placebo and had no significant change in testosterone levels. A similar study of 9 healthy men undergoing a GLP-1 RA infusion during a euglycemic clamp showed no change in free/total testosterone or luteinizing hormone (LH).[11] However, there was a decline in free/total testosterone following an oral glucose tolerance test (OGTT). A larger study[19] of 57 men showed a similar decline in testosterone and LH following an OGTT. In a separate OGTT study, this effect on testosterone and LH was present regardless of glucose tolerance.[20] Whether this acute effect has any long-term consequences on testosterone production remains unknown.

Our study contrasts with a trial of 176 men with T2DM who received metformin and either exenatide or a sulfonylurea for 3 months.[13] These participants received exenatide 5 μg twice daily for 4 weeks and then increased to 10 μg twice daily thereafter. Investigators encouraged participants to monitor blood sugars, diet, and exercise on a monthly basis. The HbA1c for both groups improved by about 1.7%. Those receiving the GLP-1 RA lost more weight (−5.7 kg) than those receiving a sulfonylurea (−0.75 kg). Additionally, total testosterone increased more in those receiving GLP-1 RA therapy (+121.72 ng/dL) than those receiving sulfonylurea (+34.67 ng/dL) but free testosterone change did not substantially differ between groups. SHBG increased more in those receiving GLP-1 RA therapy (+10.36 nmol/L) compared to those receiving sulfonylurea (+0.33 nmol/L).

SHBG is a transport molecule for testosterone; lower levels of SHBG lead to lower total testosterone levels.[21-23] Men with obesity often have lower baseline SHBG levels that rise with weight loss.[24-26] It is therefore not surprising that Shao et al. found weight loss led to a significant rise in SHBG, which is likely what caused the rise in total testosterone but not free testosterone.

While the study by Shao et al. had many study similarities to ours, the significant decrease in body weight for study participants in that study was beyond what is expected from exenatide alone. In a head-to-head trial,[27] weight loss was similar for exenatide 10 μg

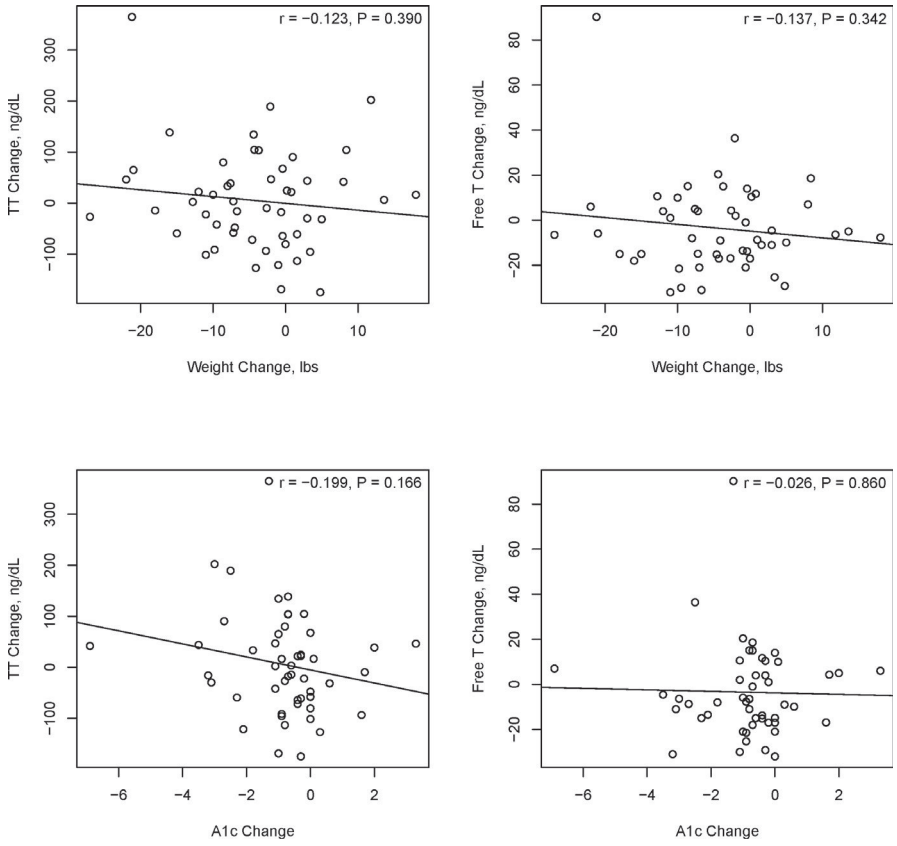

FIGURE 3 Scatter Plots of Changes in Total and Free Testosterone by Changes in Weight and A1c. There were large variations in total and free testosterone values amongst participants with weight as well as A1C changes. TT, total testosterone

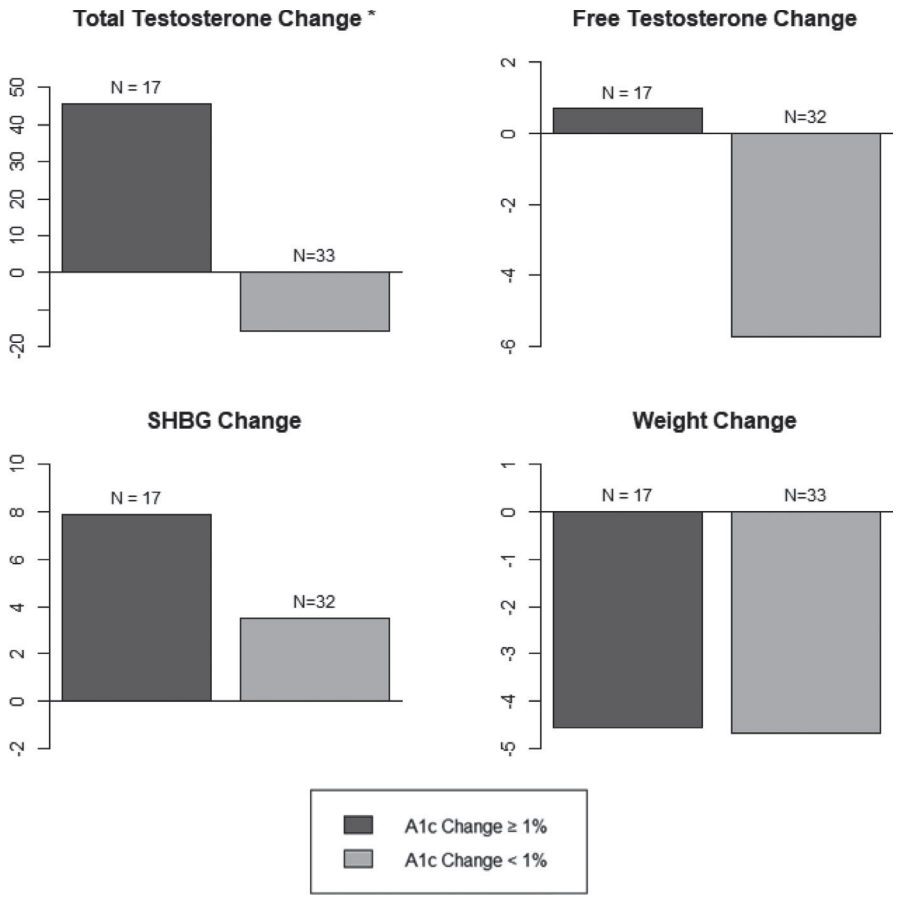

FIGURE 4 A1c Analysis. This bar graphs compares men who had A1c improvements ≥1% to those with A1c changes <1%, breaking it down by changes in total testosterone, free testosterone, SHBG, and weight. There is no evidence that changes in testosterone were related to weight loss. *Notifies a significant change for total testosterone with A1c improvements ≥1% compared to A1c changes <1% (p-value =0.04127). This relationship was not apparent for free testosterone. The change in SHBG accounted for the differences between total and free testosterone; the increase in SHBG correlated with the increase in total testosterone while the free testosterone did not change

twice daily versus exenatide ER 2 mg weekly (−1.6 to −2.8 kg vs. −2.0 to −3.7 kg, respectively). The population of Shao et al. was mostly drug naïve, and excess weight loss may have been due in part to metformin, not exenatide alone. A different study[28] showed that drug naïve patients on metformin had about a 1% weight loss at 3 months and 2% weight loss at 6 months.

We suspect the greater weight loss in the study of Shao et al. was likely multifactorial due to co-initiation of metformin along with more intensive monitoring and lifestyle interventions than in our cohort. It is possible that the population of Shao et al., being recently diagnosed with diabetes, contained more motivated individuals. Additionally, the shorter study duration likely increased dietary adherence because longer durations usually lead to higher attrition from dietary interventions. Our cohort more closely resembles real-world conditions where weight loss is generally modest, especially when patients are less likely to adhere to long-term lifestyle modifications. Had our participants achieved similar weight loss, we might have observed similar testosterone changes. Our participants lost an average of −2.27 kg while the participants in the study by Shao et al. lost an average of −5.73 kg. Moreover, their subgroup analysis showed that participants who lost >5% of their body weight had the most significant increases in serum testosterone. This is consistent with a meta-analysis on weight loss interventions that found people who lost the most weight had the greatest increase in serum testosterone.[7]

Weight loss is a relative-class effect of GLP-1 RA therapy. However, the amount of weight loss with each GLP-1 RA is variable. It is minimal with albiglutide, moderate with exenatide, and robust with semaglutide.[27] Using an alternative GLP-1 RA to exenatide ER could show greater weight loss benefits, such as liraglutide 1.8 mg daily with weight losses of −3.57 kg versus −2.68 kg [29] or, even more so, semaglutide 1.0 mg weekly with weight losses of −5.6 kg versus −1.9 kg.[30] The weight loss appears to nadir 24–30 weeks and remains relatively stable thereafter for GLP-1 RA therapy.

There were significant testosterone increases in a recent study of 30 men with obesity who received either liraglutide 3 mg daily—the treatment dose for obesity—or testosterone replacement therapy for 16 weeks.[31] They found significantly more weight loss with GLP-1 RA therapy (−7.9 kg) than with testosterone replacement therapy (−0.9 kg) ($p < 0.001$). Total testosterone significantly increased with liraglutide (+75 ± 101 ng/dL) and with testosterone replacement therapy (+170 ± 208 ng/dL); the testosterone increase in both groups was not significantly different from each other ($p = 0.239$). Sexual function improved with both therapies. They concluded that the GLP-1 RA therapy showed an overall health benefit for men with obesity and low total testosterone levels. This is an important finding in a time when many men are drawn to testosterone replacement therapy, believing it is the single best therapy for their symptoms.

Our study showed significant testosterone increases when HbA1c improvement ≥1% compared to HbA1c change <1%.The mechanism for diabetes improvement resulting in gonadal function improvement is not fully understood. In vitro studies showed that insulin stimulates the secretion of gonadotropin-releasing factors, gonadotropins, and testosterone. Hypogonadism may result because the hypothalamic–pituitary–gonadal axis does not fully respond in an insulin resistance state. Insulin resistance can lead to lower total testosterone and more frequent diagnosis of hypogonadism in men with obesity.[32] Additionally, insulin resistance and hyperinsulinemia can inhibit hepatic SHBG secretion in vitro and in vivo; SHBG influences the bioavailability of testosterone.[33] Furthermore, hyperglycemia can increase inflammation. There was a study of mice with type 1 diabetes who had progressive testicular dysfunction and a 30% reduction in testis weight compared to control mice. This effect was thought to be caused by inhibitory effects of chronic hyperglycemia mediated by testicular inflammatory cytokines.[34] It is possible that reversal of hyperglycemia improves inflammation and allows testosterone to increase. Free testosterone is the biologically active form and more consistently correlates with hypogonadal symptoms.[35] Obesity and T2DM are risk factors for hypogonadism associated with low free testosterone[36] so consideration should be given to measurement or calculation of free testosterone in this population.

Our study does have several limitations. The most significant was that as an observational study, there is no true control group. To the best of our ability, we had patients act as their own control by assessing testosterone levels before and after GLP-1 RA therapy. Participants were at liberty to make choices about diet, exercise, and sleep that could potentially impact their testosterone levels. Lifestyle education and other therapeutic recommendations were not standardized between therapeutic providers. Our pharmacy formulary limited the dosing and choice of GLP-1 RA therapy to essentially one medication. Blood draws occurred in the morning, and patients were instructed to fast, but we were not able to control for other stressors such as minor illnesses. Participant ages ranged from 36 to 73 years; they were not required to check C-peptide or GAD antibodies for diagnosis of type 2 diabetes mellitus.

Prescription of additional medication was not restricted as long as participants continued to meet inclusion and exclusion criteria. Patients were on variable combinations of diabetes medications. A majority of patients (82%) were on a DPP4i prior to study entry and then switched to a GLP1-RA, which likely blunted the HbA1c reduction. Pratley et al compared patients transitioning from sitagliptin to liraglutide with a group that was never on sitagliptin.[37] The group starting on a DPP4i had initial reduction in HbA1c by −0.9% from sitagliptin; this decreased another −0.2% to −0.5% after switching to liraglutide 1.2 and 1.8 mg/day, respectively. Final outcomes of HbA1c were similar between the liraglutide-only versus the sitagliptin-to-liraglutide group, noting there was no benefit in sequential dosing over continuous GLP1-RA therapy. In contrast, DPP4 inhibitors are weight neutral so weight loss should not be blunted when transitioning from DPP4i to GLP1-RA.[38] Pratley et al also demonstrated this with no significant weight changes on sitagliptin but then changing from sitagliptin to liraglutide 1.2 and 1.8 mg/day led to weight reductions of −1.6 and −2.5 kg, respectively.

The purpose of this study was to investigate the biochemical effects of GLP-1 RA on testosterone so we did not assess clinical symptoms of hypogonadism. As the serum testosterone level where men develop symptoms of hypogonadism may vary between individuals, this limits the clinical generalizability of our findings.

There were limitations in the statistical analysis. We included participants with baseline testosterone of various values, including predominantly men who were eugonadal. Limiting the inclusion criteria to only those with hypogonadism at baseline would likely demonstrate more profound results. The study reached its recruitment target based on power analysis, but this was still a small sample size. There was patient drop out; this was mainly due to patients either not completing the full laboratory assay prior to starting GLP-1 RA therapy or discontinuing injections because of local irritation.

Furthermore, targeted subgroup evaluation was not pre-specified in the original study protocol power analysis calculations. Therefore, our conclusions within the subgroup analysis, regarding patients with baseline total testosterone levels <320 ng/dL and HbA1c improvement ≥1%, are interesting, but would require further evaluation using appropriately powered subgroups to evaluate for significant changes. Despite the limitation, similar small subgroup analysis has been seen in previously published data.[13] Moreover, the subgroup statistics has shed light onto potential future research focuses.

Future research could investigate how much weight loss is required for GLP-1 RA therapy to increase testosterone, whether GLP-1 RA therapy alone is sufficient to treat functional hypogonadism in men with obesity and/or T2DM, and whether testosterone levels change in a dose-dependent manner similar to the weight loss effect of various DDP4 inhibitors and GLP-1 RA therapies. In addition, there is a current paucity of literature about how improvements in hyperglycemia affect testosterone and this would be worthwhile to investigate. Future studies may also benefit from comparison with a hyperglycemic drug classes like SGLT2 inhibitors that may have an effect on weight, testosterone, and glucose management to determine the true effect of GLP1-RA.

In conclusion, this study showed that men taking GLP-1 RA therapy for T2DM lost on average of 2.3 kg in 6 months and had a 0.7% improvement in HbA1c without significant changes in serum testosterone levels. Furthermore, it appears there is greater potential for increasing total testosterone in men starting at lower values and in those with more robust glucose responses to GLP-1 RA therapy.

CONFLICT OF INTEREST

The authors take public responsibility for the content of this manuscript and adhered to ethical standards. They declare that there are no conflicts of interest.

DISCLAIMER

The views expressed herein are those of the authors and do not reflect the official policy or position of Brooke Army Medical Center, the U.S. Army Medical Department, the U.S. Army Office of the Surgeon General, the Department of the Army, the Department of the Air Force and Department of Defense or the U.S. Government.

ORCID

Sky Graybill https://orcid.org/0000-0002-0556-6754

REFERENCES

1. Ponce OJ, Spencer-Bonilla G, Alvarez-Villalobos N, et al. The efficacy and adverse events of testosterone replacement therapy in hypogonadal men: a systematic review and meta-analysis of randomized, placebo-controlled trials. *J Clin Endocrinol Metab*. 2018;103(5):1745-1754.
2. Cole AP, Hanske J, Jiang W, et al. Impact of testosterone replacement therapy on thromboembolism, heart disease and obstructive sleep apnoea in men. *BJU Int*. 2018;121(5):811-818.
3. Loo SY, Azoulay L, Nie R, Dell'Aniello S, Yu OH, Renoux C. Cardiovascular and cerebrovascular safety of testosterone replacement therapy among aging men with low testosterone levels: a cohort study. *Am J Med*. 2019;132:1069-1077.
4. Vigen R, O'Donnell CI, Barón AE, et al. Association of testosterone therapy with mortality, myocardial infarction, and stroke in men with low testosterone levels. *JAMA*. 2013;310(17):1829-1836.
5. Goodman N, Guay A, Dandona P, Dhindsa S, Faiman C, Cunningham GR. AACE reproductive endocrinology scientific committee. American Association of Clinical Endocrinologists and American College of Endocrinology Position Statement on the Association of Testosterone and Cardiovascular Risk. *Endocr Pract*. 2015;21(9):1066-1073.
6. Bhasin S, Brito JP, Cunningham GR, et al. Testosterone therapy in men with hypogonadism: an endocrine society clinical practice guideline. *J Clin Endocrinol Metab*. 2018;103(5):1715-1744.
7. Corona G, Rastrelli G, Monami M, et al. Body weight loss reverts obesity-associated hypogonadotropic hypogonadism: a systematic review and meta-analysis. *Eur J Endocrinol*. 2013;168(6):829-843.
8. Casulari LA, Caldas AD, Domingues Casulari Motta L, Lofrano-Porto A. Effects of metformin and short-term lifestyle modification on the improvement of male hypogonadism associated with metabolic syndrome. *Minerva Endocrinol*. 2010;35(3):145-151.
9. Sridhar S, Walia R, Sachdeva N, Bhansali A. Effect of pioglitazone on testosterone in eugonadal men with type 2 diabetes mellitus: a randomized double-blind placebo-controlled study. *Clin Endocrinol (Oxf)*. 2013;78(3):454-459.
10. Garber A, Henry R, Ratner R, et al. LEAD-3 (Mono) study group. liraglutide versus glimepiride monotherapy for type 2 diabetes: a randomized, 52-week, phase III, double-blind, parallel-treatment trial. *Lancet*. 2009;373(9662):473-481.
11. Jeibmann A, Zahedi S, Simoni M, Nieschlag E, Byrne MM. Glucagon-like peptide-1 reduces the pulsatile component of testosterone secretion in healthy males. *Eur J Clin Invest*. 2005;35(9):565-572.
12. Giagulli VA, Carbone MD, Ramunni MI, et al. Adding liraglutide to lifestyle changes, metformin and testosterone therapy boosts erectile function in diabetic obese men with overt hypogonadism. *Androl*. 2015;3(6):1094-1103.
13. Shao N, Yu XY, Yu YM, et al. Short-term combined treatment with exenatide and metformin is superior to glimepiride combined metformin in improvement of serum testosterone levels in type 2 diabetic patients with obesity. *Andrologia*. 2018;50(7):1-9.
14. Centers for Disease Control and Prevention. *National Diabetes Statistics Report, 2020*. Centers for Disease Control and Prevention, U.S. Dept of Health and Human Services. www.cdc.gov/diabetes/pdfs/data/statistics/national-diabetes-statistics-report.

15. Niskanen L, Laaksonen D, Punnonen K, Mustajoki P, Kaukua J, Rissanen A. Changes in sex hormone-binding globulin and testosterone during weight loss and weight maintenance in abdominally obese men with the metabolic syndrome. *Diabetes Obes Metab*. 2004;6(3):208-215.
16. Kraemer HC, Thiemann S. *How many subjects? Statistical power analysis in research*. Newbury Park: Sage Publications; 1988.
17. Wu FC, Tajar A, Beynon JM, et al. Identification of late-onset hypogonadism in middle-aged and elderly men. *NEJM*. 2010;363:123-135.
18. Izzi-Engbeaya C, Jones S, Crustna Y, et al. Effects of Glucagon-like Peptide-1 on the reproductive axis in healthy men. *J Clin Endocr Metab*. 2020;105:1119-1125.
19. Iranmanesh A, Lawson D, Veldhuis JD. Glucose ingestion acutely lowers pulsatile LH and basal testosterone secretion in men. *Am J Physiol Endocrinol Metab*. 2012;302(6):E724-E730.
20. Caronia LM, Dwyer AA, Hayden D, Amati F, Pitteloud N, Hayes FJ. Abrupt decrease in serum testosterone levels after an oral glucose load in men: implications for screening for hypogonadism. *Clin Endocrinol (Oxf)*. 2013;78(2):291-296.
21. Mathur RS, Moody LO, Landgrebe SC, Peress MR, Rust PF, Williamson HO. Sex-hormone-binding globulin in clinically hyperandrogenic women: association of plasma concentrations with body weight. *Fertil Steril*. 1982;38(2):207-211.
22. Moon H, Choi I, Kim S, et al. Cross-sectional association between testosterone, sex hormone-binding globulin and metabolic syndrome: the healthy twin study. *Clin Endocrinol (Oxf)*. 2017;87(5):523-531.
23. Gyawali P, Martin SA, Heilbronn LK, et al. Cross-sectional and longitudinal determinants of serum sex hormone binding globulin (SHBG) in a cohort of community-dwelling men. *PLoS One*. 2018;13(7):e0200078.
24. Pasquali R, Vicennati V, Scopinaro N, et al. Achievement of near-normal body weight as the prerequisite to normalize sex hormone-binding globulin concentrations in massively obese men. *Int J Obes Relat Metab Disord*. 1997;21(1):1-5.
25. Van Gemert WA, Schuit AJ, van der Palen J, et al. Effect of weight loss, with or without exercise, on body composition and sex hormones in postmenopausal women: the SHAPE-2 trial. *Breast Cancer Res*. 2015;2(17):120.
26. Moran LJ, Brinkworth GD, Martin S, et al. Long-term effects of a randomised controlled trial comparing high protein or high carbohydrate weight loss diets on testosterone, SHBG, erectile and urinary function in overweight and obese men. *PLoS One*. 2016;11(9):e0161297.
27. Gentilella R, Pechtner V, Corcos A, Consoli A. Glucagon-like peptide-1 receptor agonists in type 2 diabetes treatment: are they all the same? *Diabetes Metab Res Rev*. 2019;35(1):e3070.
28. Rosenstock J, Chuck L, González-Ortiz M, et al. Initial combination therapy with canagliflozin plus metformin versus each component as monotherapy for drug-naïve type 2 diabetes. *Diabetes Care*. 2016;39(3):353-362.
29. Buse JB, Nauck M, Forst T, et al. Exenatide once weekly versus liraglutide once daily in patients with type 2 diabetes (DURATION-6): a randomised, open-label study. *Lancet*. 2013;381(9861):117-124.
30. Ahmann AJ, Capehorn M, Charpentier G, et al. Efficacy and safety of once-weekly semaglutide versus exenatide ER in subjects with type 2 diabetes (SUSTAIN 3): a 56-week, open-label, randomized clinical trial. *Diabetes Care*. 2018;41(2):258-266.
31. Jensterle M, Podbregar A, Goricar K, Gregoric N, Janez A. Effects of liraglutide on obesity-associated functional hypogonadism in men. *Endocr Connect*. 2019;8(3):195-202.
32. Saad F, Haider A, Doros G, Traish A. Long-term treatment of hypogonadal men with testosterone produces substantial and sustained weight loss. *Obesity*. 2013;21(10):1975-1981.
33. Giagulli VA, Castellana M, Murro I, et al. The role of diet and weight loss in improving secondary hypogonadism in men with obesity with or without type 2 diabetes mellitus. *Nutrients*. 2019;11(12):2975.
34. Maresch CC, Stute DC, Ludlow H, et al. Hyperglycemia is associated with reduced testicular function and activin dysregulation in the $Ins2^{Akita+/-}$ mouse model of type 1 diabetes. *Mol Cell Endocrinol*. 2017;5(446):91-101.PMID: 28214591.
35. Antonio L, Wu FC, O'Neill TW, et al. Low free testosterone is associated with hypogonadal signs and symptoms in men with normal total testosterone. *J Clinic Endocrinol Metabol*. 2016;101(7):2647-2657.
36. Dhindsa S, Miller MG, McWhirter CL, et al. Testosterone concentrations in diabetic and nondiabetic obese men. *Diabetes Care*. 2010;33(6):1186-1192.
37. Aroda VR, Henry RR, Han J, et al. Efficacy of GLP-1 receptor agonists and DPP-4 inhibitors: meta-analysis and systematic review. *Clin Ther*. 2012;34(6):1247-1258.
38. Pratley RE, Nauck MA, Bailey T, et al. 1860-LIRA-DPP-4 study group. Efficacy and safety of switching from the DPP-4 inhibitor sitagliptin to the human GLP-1 analog liraglutide after 52 weeks in metformin-treated patients with type 2 diabetes: a randomized, open-label trial. *Diabetes Care*. 2012;35(10):1986-1993.

How to cite this article: Graybill S, Hatfield J, Kravchenko M, et al. Neutral effect of exenatide on serum testosterone in men with type 2 diabetes mellitus: A prospective cohort. *Andrology*. 2021;9:792–800. https://doi.org/10.1111/andr.12966