Injection of Testosterone May be Safer and More Effective than Transdermal Administration for Combating Loss of Muscle and Bone in Older men

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Focus: The focus of this review focuses on musculoskeletal effects of testosterone and the place of these benefits in the discussion of risk-benefit ratio for testosterone replacement therapy.

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Abstract

The value of testosterone replacement therapy (TRT) for older men is currently a topic of intense debate. While US testosterone prescriptions have tripled in the last decade (9), debate continues over the risks and benefits of TRT. TRT is currently prescribed for older men with either low serum testosterone (T) or low T plus accompanying symptoms of hypogonadism. The normal range for serum testosterone is 300 to 1000 ng/dL. Serum T ≤ 300 ng/dL is considered to be low and T ≤ 250 is considered to be frank hypogonadism. Most experts support TRT for older men with frank hypogonadism and symptoms. Treatment for men who simply have low T remains somewhat controversial. TRT is most frequently administered by i.m. injection of long-acting T esters or transdermally via patch or gel preparations and infrequently via oral administration (see Table 1). TRT produces a number of established benefits in hypogonadal men, including increased muscle mass and strength, decreased fat mass, increased bone mineral density and improved sexual function and in some cases those benefits are dose-dependent. For example, doses of TRT administered by i.m. injection are typically higher than those administered transdermally, which results in greater musculoskeletal benefits. TRT also produces known risks including development of polycythemia (hematocrit > 50) in 6% of those treated, decrease in HDL, breast tenderness and enlargement, prostate enlargement, increases in serum PSA and prostate-related events and may cause suppression of the hypothalamic-pituitary-gonadal axis. Importantly, TRT does not increase the risk of prostate cancer. Putative risks include edema and worsening of sleep apnea. Several recent reports have also indicated that TRT may produce cardiovascular (CV) risks, while others report no risk or even benefit. To address the potential CV risks of TRT, we have recently reported via meta-analysis that oral TRT increases CV risk and suggested that the CV risk profile for i.m. TRT may be better than that for oral or transdermal TRT.

Scope of the review. Herein, we review the literature which indicates that i.m. injected TRT produces greater musculoskeletal benefits and lower cardiovascular risk compared to
transdermal TRT. TRT also produces risk of polycythemia, prostate enlargement and

suppression of the hypothalamic-pituitary-gonadal axis. The effects of injection vs. transdermal

administration on these risks are unknown. We also review the literature discussing the use of

5α-reductase inhibitors as a promising means of improving the safety profile of TRT.
Definition of hypogonadism

The Endocrine Society recommends TRT for men with androgen deficiency, defined as low serum T with consistent symptoms and signs of hypogonadism (5), including decreased sexual function, loss of axillary and pubic hair, low bone mineral density, loss of motivation, mood or concentration and loss of muscle strength and work capacity. In older men, hypogonadism is commonly defined as a serum total T concentration of ≤ 300 ng/dL (i.e. below the normal range) or ≤ 250 ng/dL (i.e. frank hypogonadism). Both groups may benefit from TRT. Determination of serum T should be based on the average of 2 blood samples drawn before 10:00 AM. Kaufman and Vermeulen (45) have reviewed the literature and reported that approximately 20% of men over the age of 60 have a serum total T concentration of ≤ 320 ng/dL. Similarly, we have reported (19) that 24% of men over 60 have a serum total T of ≤ 300 ng/dL. Additionally, hypogonadism is sometimes defined as low free T or low bioavailable T (bioT). In this regard, a small fraction of circulating T is unbound while the bulk is either loosely bound to albumin or very tightly bound to steroid hormone binding globulin (SHBG). BioT is the sum of the free and albumin-bound T and represents the most physiologically important T fraction. BioT is the fraction able to cross cell membranes and bind to androgen receptors (ARs). An increase in SHBG due to aging (85), traumatic events, such as spinal cord injury (11) will result in reduced bioT. Total T assays are available to most clinicians, while free T and bioT assays are less available. However, serum bioT may be estimated from serum total T, SHBG and albumin using one of several empiric formulas (57, 86). The Endocrine Society recommends that treatment be individualized including a determination of whether the patient has primary hypogonadism (primary deficit lies within the testes) or secondary hypogonadism (primary deficit lies outside of the testes) (13). When T-enanthate or T-cypionate are injected, one should aim for a serum total T concentration of 400 – 700 ng/dL at one week after the most recent injection.

Modes of TRT and doses delivered
Testosterone use in the United States increased more than 3-fold between 2000 and 2011 (36). The most common modes of T administration are patch and gel preparations and i.m. injection of long acting T esters. Transdermal doses are typically intended to be replacement doses with the patch being administered at a doses of 5-10 mg T/day and gel administration involving a somewhat greater amount of T due to low absorption (see Table 1). Injection of T-esters typically delivers higher amounts, 50 to 400 mg every two to 4 weeks. Esters such as T-enanthate or T-cypionate have virtually no aqueous solubility and remain in the muscle depot until muscle esterase activity releases T, which can then enter the circulation. Administered T may be converted to estradiol via the action of aromatase or to dihydrotestosterone (DHT) by the action of 5-α reductase (see Figure 1), both of which exert biologic actions at estrogen receptor and ARs, respectively. As such, T may be considered both a hormone (because it binds to ARs) and a prohormone for the synthesis of estradiol and DHT. Interestingly, the mode of TRT administration appears to alter the metabolism of T, as evidenced by our recent meta-analysis (18) reporting that transdermal T (patch and gel) elevated serum DHT 5.46 fold, while i.m. injected T elevates serum DHT only 2.2 fold. This surprising phenomenon occurs despite the fact that transdermal and i.m. TRT elevated serum T to a roughly similar degree and may be explained by relatively high expression of 5-α reductase in skin (40) vs. lower expression in skeletal muscle (93).

Effect of TRT on muscle, bone and body composition

Age is accompanied by a progressive decline in muscle mass with a decrease in both the number and diameter of muscle fibers, especially type II (fast) fibers (59). Fiber loss with aging is secondary to a loss in motor neurons (81). TRT increases muscle mass and strength by increasing the cross-sectional area of both type I and type II fibers in a dose-dependent manner (70). At low replacement doses, T administration produces muscle protein accretion mainly by preventing protein degradation (30), which is mediated, at least in part, by an increase phosphorylation-induced inactivation of FOXO3a (89). At higher doses, T also stimulates
muscle protein synthesis via a number of mechanisms that include proliferation of muscle
satellite cells and donation of their nuclei to the myofibril (71), elevation of muscle IGF-I (69) and
activation of the Akt/mammalian target of rapamycin (mTOR) pathway (90). Binding of T to
ARs present in mesenchymal pluripotent cells causes translocation of β-catenin to the nucleus,
causing them to differentiate into the myocyte lineage rather than the adipocyte lineage (68).
Preservation of lower body muscular strength is an important factor in maintaining
independence in older individuals. Resistance training produces substantial strength increases
in the elderly. For example, Villanueva et al. found that in a group of healthy older men,
resistance training for 12 weeks increased 1-RM leg strength by 100% (88). By contrast, TRT
produces smaller gains, but does so independently of exercise. In general, the response to
lower doses of TRT administered transdermally may be considered modest, while the effects on
injected TRT are moderate. Transdermal TRT to older men often increases lean mass,
however most studies do not report increases in strength (20, 48). A 2006 meta-analysis by
Ottenbacher et al. (60), assessing 11 RCT of TRT in older men, reported that injected TRT
produced a “moderate” increase in muscle strength, while the effects of transdermal and oral
TRT were much less. We surveyed 10, mostly more recent, RCTs of TRT in older men lasting
12 weeks or more and reporting 1-RM strength as an outcome. None of the four transdermal
studies (10, 37, 47, 58) reported a significant increase in 1-RM strength. In contrast, all six of
the i.m. injection studies (19, 22, 30, 64, 75, 78) reported significantly increased strength. We
administered a moderately high dose of 125 mg/week T-enanthate i.m. to older hypogonadal
men and observed increases of 8-14% in 1-RM strength over a 12-month period, with most of
the improvement occurring during the first 3 months (19). Storer et al. (75) administered a
higher dose of 300 mg T/week i.m. to young eugonadal men for 5 months and reported that 1-
RM leg press strength was increased 23% and leg press power by 39%. The benefits with
even higher doses of i.m. T (300 to 600 mg/week) are even more substantial (upwards of 20%
increase in 1-RM strength in older men over 20 weeks), but the number of adverse events
observed at these doses preclude their clinical implementation (14). Thus injected TRT has value for increasing muscle strength in older men, especially because not all patients have the means or ability to exercise. In addition, hypogonadism may reduce both the response to and the motivation for resistance exercise.

Effect of TRT on bone

Men over the age of 65 are subject to an increased incidence of osteoporosis and to increased falls and fractures ultimately contributing to increased mortality (15). In older men, low serum T is associated with osteopenia (49) and increased fracture risk (55). T administration increases BMD, mainly by suppressing bone resorption (12, 16). TRT may increase BMD in men by a direct AR-mediated effect of T or by an indirect action requiring conversion to estradiol (94). In this regard, low serum estradiol is more strongly associated with osteopenia in older men than is low serum T (49). Interestingly, the indirect effects of T may be more important than the direct effects, as evidenced by the work of Falahati-Nini et al. (28), who observed elevated blood markers of bone resorption following combined administration, to older men, of a GnRH agonist and an aromatase inhibitor to inhibit production of T and estradiol, respectively. Subsequent administration of transdermal estradiol alone suppressed markers of bone resorption, while transdermal T alone did not, while full suppression of bone resorption occurred only with combined T plus estradiol administration. While bone protection in response to replacement doses of T requires aromatization, bone protection resulting from high doses of androgen does not appear to.

We have reported that i.m. administration of either T or its non-aromatizable analog trenbolone can completely prevent orchiectomy-induced bone loss in skeletally mature rats (54), and that co-administration of the aromatase inhibitor anastrozole does not inhibit the effects of T and trenbolone (12).

In older men, T increases BMD in regions that have a large component of cancellous bone, such as the lumbar spine and hip (84). These increases are important because they occur at
sites where fractures frequently occur in the elderly. We have reported that older men receiving 125 mg T enanthate/week i.m. for 1 year exhibited a 4% increase in lumbar spine BMD and a 2% increase in hip BMD over 12 months (19). Amory et al. have reported continued BMD improvement through 36 months of i.m. TRT with lumbar spine BMD increasing 10% and hip BMD increasing 3% compared to baseline (7). Interestingly, in a 2006 meta-analysis of 8 TRT trials in older hypogonadal men, Tracz et al found that i.m. TRT produced a significant 8% increase in lumbar spine BMD and a non-significant 4% in hip BMD, while transdermal TRT produced no increases in BMD (84). We have also reported that in rats, T-enanthate prevents orchiectomy-induced loss of bone mechanical strength (92). The large clinical trials needed to assess fracture risk following TRT have not yet been conducted.

Adverse effects of TRT

Established risks of TRT. Meta-analysis has confirmed 3 adverse events (AEs) resulting from TRT (21, 29, 34) 1) polycythemia occurring in ~6% of participants 2) an increased number of prostate-related events, and 3) a small reduction in HDL cholesterol. Prostate events consist of the combined incidence of elevated PSA, prostate biopsy necessitated by results of digital rectal exam, increased urinary symptoms, and prostate cancer. Meta-analysis by Calof et al. (21) reported no evidence that T administration increases prostate cancer (odds ratio = 1.09 with no trend toward significance), when considered as an independent outcome. More recently, a 15-year retrospective study of 150,000 men by Kaplan and Hu (43) found that TRT was not associated with prostate cancer. In addition, TRT inhibits endogenous T production and suppresses the hypothalamic-pituitary-gonadal axis. Endogenous T production may not resume or may be diminished following cessation of TRT. The relative effects of i.m. injected vs. transdermal TRT on prostate enlargement, polycythemia and suppression of the hypothalamic-pituitary-gonadal axis are unknown.

In addition, some risks depend specifically on the mode of TRT administration (13). Injected TRT may cause pain or bleeding at the site of injection and should not be given to men
receiving anti-coagulants. Patches may cause skin reactions and gel may result in transfer of testosterone to a partner.

**Less common and putative risks of TRT.** Testosterone may also cause edema, breast tenderness and gynecomastia (79), effects that are thought to result from elevation of estradiol subsequent to TRT. Because adipose tissue is the principal site of systemic aromatization (conversion of T to estradiol), TRT is often contraindicated for men with a BMI of 30 or more.

Causing or worsening of sleep apnea is frequently listed among potential AEs of TRT, based primarily on case reports in the literature. Meta-analysis of 19 clinical trials through 2005 by Calof et al. (21) showed no significant increase obstructive sleep apnea with TRT, however the studies were not conducted with polysomnography. The one study using this technique was performed by Hoyos et al. (39), who treated middle aged men who had severe obstructive sleep apnea with T. T mildly worsened sleep-disordered breathing after 7 weeks of treatment, but not after 18 weeks.

**Possible CV risks of TRT.** Recently, 4 reports have caused concern regarding the potential CV risks of testosterone administration. Basaria et al. (10), in their randomized controlled trial of T gel administration, reported a greater number of CV AEs in treated vs, placebo, resulting in cessation of the trial. However, it should be noted that the trial was not designed to assess pre-specified CV surrogate outcomes or clinical endpoints. Furthermore, the increased CV-related adverse events were considered as a composite endpoint including events of varying severity and mechanisms. Vigen et al. (87), in a retrospective study of men with low T and angina who also had undergone angiography, reported a higher CV risk in those who subsequently received T than in those who did not. Methodology of this paper was criticized in numerous letters to the editor of JAMA. Another observational study by Finkle et al. (31) reported a greater risk of myocardial infarction in men who had received a prescription for T. Finally, Xu and colleagues (91) published a 2013 meta-analysis of CV adverse events in 27 randomized controlled trials (RCTs) of TRT published through December of 2012 and reported a statistically significant odds
ratio (OR) of 1.54, indicating that participants receiving TRT were 54% more likely to develop CV AEs. As a result of these studies, the FDA (4), VA (1) and Endocrine Society (2) have all issued advisories calling for more research on potential TRT-related CV risks. The latest FDA Drug Safety Communication, issued March 2015, will require manufacturers of approved testosterone products to conduct clinical trials assessing risks of heart attack and stroke (3).

We have recently performed the largest meta-analysis of RCTs to date (including 35 RCTs of TRT lasting 12 weeks or more, reporting CV adverse events and published through May of 2014) evaluating TRT-related CV risks (18) and using guidelines for analysis of low-frequency events (95). Our main finding was that the form of testosterone (18) administration influenced its CV risk profile (see Table 2). Specifically, we reported the following new and significant findings. Because Xu et al. did not use statistical techniques suited to analysis of low-frequency events and because some recent studies have reported fewer TRT-induced CV AEs, the estimate of risk for CV AEs is revised downward (RR = 1.28, non-significant). Oral TRT produces significant risk for CV AEs (RR = 2.20, p = 0.015). Transdermal (patch or gel) TRT produces a non-significant directional trend toward CV risk (RR = 1.27) and i.m. TRT produces a non-significant directional trend toward CV protection (RR = 0.66). Transdermal and oral TRT cause greater elevation of serum DHT (but not T) compared to injected TRT (see Table 1).

Serum DHT concentrations following transdermal and oral TRT correspond to the concentrations that have been linked to CV disease and mortality in observational studies (66).

Corona et al. (25) have published a new meta-analysis of 75 studies, assessing the CV risks of TRT, using less stringent inclusion criteria compared to our study and that of Xu et al. Corona et al. found no risk of all CV events (odds ratio = 1.01) or of serious CV events (odds ratio = 1.07).

Possible CV benefits of testosterone.

It is well-described that low serum T is associated with poor CV outcomes including coronary artery disease, heart failure and stroke (82). In particular, Shores et al. have reported
that low serum T in men is associated with increased incidence of CV disease and increased
all-cause mortality (66). In addition, several studies directly demonstrate CV benefits of TRT.
For example, English et al. (27) have shown that, in men with stable angina, low-dose TRT (5
mg/day by patch) for 12 weeks caused a significant 17% increase in time to the development of
ischemic EKG changes (i.e. 1-mm ST segment depression) during treadmill exercise testing.
Stout et al. (76) have shown that TRT in men with chronic heart failure improves exercise
performance (increased VO2max). Toma et al. (83) published a meta-analysis of 4 studies
showing that TRT improved exercise capacity in heart failure patients. TRT-induced CV
benefits may derive from the properties of T as a coronary dilator. Chou et al. (24) reported that
in dogs, injection of T into the coronary circulation caused vasodilation and increased coronary
blood flow and we have reported that T-enanthate improves recovery of aortic flow, cardiac
work and left ventricular developed pressure in an orchiectomized male rat model of global
ischemia/reperfusion (17).

The role of DHT in responses to testosterone

As previously discussed, 5α-reductase enzyme actively converts T to DHT in a local tissue-
specific manner, which local increases in action. As such, it remains biologically and clinically
important to evaluate the role of 5α-reductase in mediating the effects of TRT, especially given
that DHT binds to ARs with approximately three times the affinity of T (93) and that DHT may
mediate several of the AEs resulting from TRT. 5α-reductase exists in 3 isoforms (type I, II and
III), with skeletal muscle expressing type I and III, but not II (93). We have shown that in older
hypogonadal men, finasteride, a specific inhibitor of 5-α reductase type II, did not block T-
induced increases in lean mass or muscle strength (19). Similarly, Bhasin et al. have shown in
young men, that dutasteride, a dual inhibitor of 5-α reductase type I and II, also does not inhibit
T-induced increases in lean mass and 1-RM strength. In addition, both our laboratory (19) and
others (7) have shown that DHT is not required for the T-induced increase in BMD or hematocrit
(62). Macukat et al. have shown that dutasteride administration does not decrease, BMD in
older men (51). Taken together, these studies suggest that conversion of T to DHT is not required for the effects of TRT on muscle, bone or hematocrit. In contrast, some evidence suggests that DHT may cause the putative risks resulting from TRT. As discussed above, the higher circulating DHT levels, obtained with transdermal as opposed to injected T, may be responsible for the trend for increased CV risk with transdermal TRT. In observational studies, Shores et al have shown that high DHT is associated with increased cardiovascular events (66) and increased incident ischemic stroke (65). Interestingly, Zwadlo et al. (96) have shown that cardiac expression of 5-α reductase is markedly increased in humans with heart failure and in a mouse aortic-constriction model of heart failure. In the mouse model, treatment with finasteride attenuates cardiac hypertrophy and improves left ventricular function. Similarly, Rubio-Gayosso et al. (63) reported that T-administration protects against cardiac ischemia/reperfusion (I/R) injury in male rats, and that inhibition of 5α-reductase reduces I/R injury in both orchiectomized and intact rats, while DHT administration worsens I/R damage. Taken together, this evidence suggests that conversion of T to DHT may underlie CV AEs resulting from TRT.

Combination therapy of T plus a 5-α reductase inhibitor

The observation that DHT may mediate several AEs resulting from TRT but is not required for musculoskeletal benefits, results in the proposed addition of finasteride or dutasteride to improve the safety of TRT. In our study of 1 year of i.m. TRT in older men, T alone caused a 40% increase in prostate volume, while T + finasteride produced the same musculoskeletal benefits as T alone, but with no prostate enlargement (19). Similarly, Amory et al. (7) reported that T + finasteride produced less prostate enlargement than did T alone, and did not inhibit T-induced increases in BMD, although T-induced increases in muscle strength were not observed in this study. In a trial of 18,882 men, Thompson et al. demonstrated that finasteride reduced the incidence of all prostate cancer by 30%, while increasing the incidence of high-grade prostate cancer by 17% (81). However, this study may have underestimated the benefit of
finasteride because finasteride shrinks the prostate, making detection of prostate cancer easier.

Taken together, these findings indicate that co-administration of finasteride with T may increase its safety without sacrificing benefits.

Summary and recommendations

For treatment of older hypogonadal men, there are advantages to administering TRT by injection, rather than transdermally or orally. First, the musculoskeletal benefits are greater, due to the higher doses administered i.m. vs transdermally. Second, although the doses are higher, i.m. TRT may not pose the same CV risks that result from transdermal TRT. A possible explanation for the latter phenomenon is that transdermal T causes greater elevation of serum DHT, due to significant expression of 5α-reductase in skin, but not in muscle. Meta-analysis of existing randomized placebo-controlled trial is, to date, insufficient to definitively assess the CV effects of TRT. However, existing data exhibit trends indicting 1) that TRT may not accelerate underlying early-stage prostate cancer 2) that transdermal TRT may cause CV risk and 3) that i.m. injected TRT may cause CV benefit. In addition, several studies demonstrate that in older hypogonadal men, the combination of i.m. T plus finasteride produces musculoskeletal benefits without the prostate enlargement that results from T alone (7, 19). Finasteride produces relatively few adverse events and may also produce cardiovascular benefits and/or reduce prostate cancer by reducing DHT. While further research is needed, it appears at this time that i.m. injected T plus finasteride may be both the safest and most effective treatment for older hypogonadal men.
Figure 1. T may act directly at androgen receptors or indirectly following conversion to estradiol or DHT.
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<td>i.m. injection of T-cypionate or t-enanthate</td>
<td>Depot-Testosterone®</td>
<td>weekly or biweekly</td>
<td>50-400 mg T/every 2 to 4 weeks</td>
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<td>Andriol®</td>
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Table 1. Common modes of testosterone administration
Table 2. Risk of CV events in placebo-controlled randomized clinical trials published through May 2014. CV risk varies by route of T administration. Oral TRT produces significant risk. Gel and Patch TRT produce possible risk, while i.m. injected TRT produces possible benefit (18).
1. Department of Veterans Affairs VHA Pharmacy Benefits management Advisory panel: 
Testosterone Products and Cardiovascular Safety
2. Endocrine Society Statement on The Risk of Cardiovascular Events in Men Receiving Testosterone Therapy
3. FDA Drug Safety Communication: FDA cautions about using testosterone for low testosterone due to aging; requires labeling change to inform of possible increased risk or heart attack or stroke with use. 2015.
4. Testosterone Products: Drug Safety Communication - FDA Investigating risk of Cardiovascular Events
5. Testosterone Therapy in Adult Men with Androgen Deficiency Syndromes: an Endocrine Society Clinical Practice Guideline


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90. Wu Y, Bauman WA, Blitzer RD, and Cardozo C. Testosterone-induced hypertrophy of L6 myoblasts is dependent upon Erk and mTOR. *Biochemical and biophysical research communications* 400: 679-683, 2010.


Figure 1. T may act directly at androgen receptors or indirectly following conversion to estradiol or DHT.