Androgen Supplementation in Older Women: Too Much Hype, Not Enough Data

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Androgen supplementation in women has received enormous attention in the scientific and lay communities. That it enhances some aspects of cognitive function, sexual function, muscle mass, strength, and sense of well-being is not in question. What is not known is whether physiological testosterone replacement can improve health-related outcome in older women without its virilizing side effects. Although it is assumed that the testosterone dose-response relationship is different in women than in men and that clinically relevant outcomes on the above-mentioned effects can be achieved at lower testosterone doses, these assumptions have not been tested rigorously. Androgen deficiency has no clear-cut definition. Clinical features may include impaired sexual function, low energy, depression, and a total testosterone level of less than 15 ng/dL, the lower end of the normal range. Measurement of free testosterone is ideal, because it provides a better estimate of the biologically relevant fraction. It is not widely used in clinical practice, because some methods of measuring free testosterone assay are hampered by methodological difficulties. In marked contrast to the abrupt decline in estrogen and progesterone production at menopause, serum testosterone is lower in older women than in menstruating women, with the decline becoming apparent a decade before menopause. This article reviews testosterone's effects on sexual function, cognitive function, muscle mass, body composition, and immune function in postmenopausal women. **J Am Geriatr Soc 50:1131–1140, 2002.**

Key words: androgen; menopausal women; cognitive; sexual; muscle mass; bone density

That testosterone supplementation might improve some aspects of cognitive and sexual functions, muscle mass and strength, bone mineral density, and sense of well-being is not in question, but it is not known whether physiological testosterone replacement can induce clinically meaningful improvements in health-related outcomes in older women without its limiting, virilizing side effects. It has been assumed that testosterone dose-response relationships are different in women than in men and that clinically significant effects on psychosexual function, body composition, physical function, bone mineral density, and other health-related outcomes can be achieved at testosterone doses and concentrations that are substantially lower than those required to produce similar effects in men. Neither of these assumptions has been tested rigorously. Furthermore, the premise that the organ systems that are the targets of virilizing side effects, such as the skin, hair, vocal cords, and clitoris, differ from muscle and bone in their testosterone sensitivity remains unsubstantiated. The clinical applications of testosterone in women are critically predicated upon the postulate that, by appropriate selection of testosterone dose, clinically beneficial effects can be dissociated from virilizing side effects.

There is enormous public interest in and media fascination with the issue of androgen supplementation in women. For instance, in 2000, stories related to this topic appeared in many major U.S. newspapers and on the Oprah Winfrey show and other television network programs in the United States, Australia, and Europe. The number of stories appearing in the lay press in the past 2 years has far exceeded the number of randomized clinical trials.

In spite of the growing media attention, the issue of androgen supplementation in women has remained controversial in the scientific community. Many uncertainties have contributed to a lack of consensus. The commercially available assays for total and free testosterone were developed for the measurements of much higher circulating concentrations in men; these assays have generally lacked the sensitivity and precision required to accurately measure the lower levels of testosterone in older women.[1] There has been a paucity of normative data on testosterone levels in menstruating women, older women, and women with chronic illnesses; this has made it difficult to define androgen deficiency in women in precise quantitative terms. The available formulations for androgen administration were

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developed for the replacement of the much higher doses required for the treatment of hypogonadal men. Few pharmacokinetic data exist on the bioavailability and clearance of androgens delivered from the available formulations in women. Therefore, it is not surprising that many previous clinical studies in women used pharmacological doses of testosterone in relatively unphysiological experimental paradigms. The objective of physiological testosterone replacement is to restore serum total and free testosterone concentrations to a range that is mid-normal to high normal for healthy younger women. Testosterone regimens that increase serum testosterone levels into the supraphysiological range should be viewed as pharmacological.

Sexual dysfunction in women, a highly complex, multifactorial issue, has become synonymous with androgen deficiency in the lay press. Observations that pharmacological doses of testosterone might improve sexual function in subsets of women with sexual dysfunction have been unjustifiably extrapolated to advocate testosterone replacement as a general treatment for sexual dysfunction in older women.

It would be incorrect to assert that testosterone supplementation of older women has no role in clinical practice, but the available data do not warrant a general recommendation for testosterone replacement for all postmenopausal women.

BIOLOGY OF TESTOSTERONE PRODUCTION IN MENSTRUATING, POSTMENOPAUSAL, AND OLDER WOMEN

Table 1 lists the adrenal androgens and their potencies and concentrations in serum for premenopausal and postmenopausal women.

Adrenal and ovarian production of androgens in healthy younger menstruating women collectively contributes to secretion of approximately 300 μg testosterone daily into the general circulation.[2] Approximately half of the circulating testosterone is derived from ovarian secretion.[3] The adrenal gland produces testosterone precursors dehydroepiandrosterone (DHEA), dehydroepiandrosterone sulfate (DHEAS), and androstenedione, whose peripheral conversion to testosterone contributes the other half of circulating testosterone.[4] Although the current dogma assumes approximately equal contribution from the ovaries and the adrenal glands, these assumptions may not be entirely correct. The data from studies in which testosterone production rates were measured after suppression of the hypothalamic-pituitary-adrenal axis with dexamethasone administration are suspect because dexamethasone is known also to suppress ovarian steroidogenesis. DHEAS is secreted entirely from the adrenal gland, whereas the adrenal glands and the ovaries secrete DHEA. The majority of circulating DHEA is derived from peripheral conversion of DHEAS. In women of all ages, the metabolism of DHEA and DHEAS into bioactive sex steroids may occur in many tissues, including adipose, bone, muscle, prostate, and breast tissue and the skin, the brain, the ovary, the testes, and the liver. DHEA and DHEAS are enzymatically converted to testosterone and then to DHT in the adrenal glands, ovaries, and other peripheral tissues. Circulating androstenedione is derived from secretion by zona fasciculata of the adrenal glands and stromal cells within the ovaries.

Androgen Levels During The Menstrual Period

In regularly ovulating women, plasma levels of testosterone and androstenedione rise gradually during the follicular phase to reach their highest levels in the preovulatory phase, with a second rise in androstenedione during the late luteal phase.[1,5] Serum testosterone concentrations during the mid-follicular phase are not significantly different from those in mid-luteal phase.[1] Ovariectomy causes a drop in serum testosterone and androstenedione by about 50% each.[6]

Changes In Androgen Levels with Menopause and Aging

Serum testosterone levels are lower in older women than in younger menstruating women. The decline in DHEAS and testosterone becomes apparent in the decade before menopause and is gradual and progressive[7] such that the testosterone level of women in their 60s is about 50% that of women in their 20s.[8] This is in contrast to the dramatic decline in estradiol and progesterone production that occurs at menopause. The progressive decline of DHEAS and testosterone with age is independent of the menopausal transition.[8] Although some studies have reported a decrease in androstenedione concentrations during menopause,[9] a large cross-sectional epidemiological study in Australia reported no significant change in serum total testosterone concentrations in the perimenopausal period, demonstrating that the ovarian androgen secretion is not attenuated in most women at menopause.[10] It has been reported that some ovaries may undergo stromal hyperplasia

Table 1. Relative Androgenic Activity and Levels of Adrenal Androgens

Steroid	Androgenic Activity*	Normal Serum Levels in Premenopausal Women†	Normal Serum Levels in Postmenopausal Women†
Dihydrotestosterone	300	4–22 ng/dL	3–20 ng/dL
Testosterone	100	10–55 ng/dL	7–40 ng/dL
Androstenedione	10	60–245 ng/dL	30–120 ng/dL
DHEA	5	350–700 ng/dL	150–300 ng/dL
DHEAS	5	20–250 μg/dL	10–150 μg/dL

*Relative activity based on testosterone = 100.
†Representative normal values, actual values will depend on assay and laboratory used.
DHEA = dehydroepiandrosterone; DHEAS = dehydroepiandrosterone sulfate.

under the control of elevated gonadotropins[9] and produce even higher amounts of androgens than they produced before menopause. Nevertheless, on average, testosterone production continues to decrease gradually after the fourth decade.[11]

Testosterone Metabolism

Testosterone serves not only as an androgenic hormone, but also as a prohormone; it is converted in the periphery into two active metabolites, estradiol and dihydrotestosterone (DHT). Testosterone's effects on the skin require its obligatory 5-α-reduction to dihydrotestosterone.[12] In contrast, testosterone's effects on bone resorption, gonadotropin suppression, plasma lipids, brain organization, and some aspects of cognitive function require its aromatization to estradiol.[13] We do not know whether 5-α-reduction of testosterone to DHT is obligatory for mediating its effects on the muscle and cortical bone formation.

Labrie et al.[14] have suggested that serum concentrations of testosterone and DHT may underestimate total androgenic activity and that conjugated metabolites of DHT and testosterone, such as androsterone glucuronide, androstane-3α,17β-diol-glucuronide, androstane-3β,17β-diol-glucuronide and androsterone-sulfate, may be more reliable markers of the androgen action at the end organ.[15,16] We do not know whether measurement of 3-α androstanediol glucuronide would provide a better marker of androgen action than serum levels of total and free testosterone.

MEASUREMENT OF TOTAL AND UNBOUND TESTOSTERONE

In healthy women, approximately 50% to 60% of testosterone is bound to sex hormone binding globulin (SHBG) and 30% to 40% to albumin; only 0.5% to 3% is unbound. The free hormone hypothesis assumes that only the free and loosely bound (testosterone bound to albumin) exerts biologic effects.[17] Many factors affect SHBG; thyroid hormone, estrogen, and aging increase it and testosterone, glucocorticoids, growth hormone, and insulin decrease it.[18] Although measurements of unbound testosterone should provide a better estimate of the biologically relevant fraction, in practice, some measurements of free and bioavailable testosterone have been hampered by methodological difficulties, particularly in women. The commercially available assays for the measurement of unbound testosterone include the equilibrium dialysis method, the bioavailable testosterone by the ammonium sulfate precipitation method, free testosterone indices calculated from the measured total testosterone and SHBG concentrations, and tracer analog methods for estimates of free testosterone. Of these methods, the equilibrium dialysis method for the measurement of unbound testosterone and the ammonium sulfate precipitation method for the measurement of albumin-bound and unbound testosterone (bioavailable testosterone) are acceptable methods that have good clinical correlation and are accurate, independent from the effects of SHBG concentrations, and available at specialized commercial endocrinology laboratories. The estimates of free testosterone, calculated from total testosterone and SHBG concentrations, have also been shown to correlate with free testosterone concentrations measured by dialysis in men. These algorithms have not been extensively tested in women.[19] Tracer analog methods of measuring free testosterone are widely available but are affected by SHBG concentrations and do not provide an accurate measure of unbound testosterone[19] and are not recommended. A recent commentary by Rosner emphasized that direct radioimmunoassay of free testosterone by the tracer analog method may underestimate its concentration.[20]

ANDROGEN DEFICIENCY STATES IN WOMEN

There is no consensus on a clinical or biochemical definition of androgen deficiency in women. A physical and behavioral symptom complex termed "female androgen deficiency syndrome" includes impaired sexual function, loss of energy, and depression.[21] Based on the distribution of serum total and free testosterone concentrations in healthy menstruating women, androgen deficiency could be defined by serum total testosterone concentrations less than 15 ng/dl, the lower end of the normal female range.[1]

The causes of androgen deficiency in women can be divided into ovarian, adrenal, central, and systemic causes. Ovarian causes include premature ovarian failure, Turner's syndrome, and surgical or chemical ovariectomy. Gonadal dysgenesis; streak gonads; estrogen deficiency; and low circulating levels of androstenedione, testosterone, free testosterone, and SHBG characterize Turner's syndrome.[22] Most of these women are receiving estrogen and progesterone replacement, which further decreases their free androgen levels by increasing the SHBG concentrations; in addition, luteinizing hormone suppression by hormone replacement therapy (HRT) may further decrease the stimulus for ovarian androgen production.[19] It is possible that the reduction in free androgen levels induced by traditional HRT might adversely affect sexual function in postmenopausal women.

Primary adrenal insufficiency is associated with deficiencies of glucocorticoids and adrenal androgens. Central causes of androgen deficiency include disorders affecting the pituitary or the hypothalamus. Panhypopituitarism affects androgen secretion from adrenal and ovarian sources; not surprisingly, patients with panhypopituitarism have lower circulating concentrations of total and free testosterone and androstenedione than those found in patients with adrenal or ovarian failure alone.[23] Gonadotropin-releasing hormone agonist or antagonist analogs, often used for treating endometriosis and other reproductive disorders, suppress gonadotropin secretion. Glucocorticoid therapy suppresses corticotropin-releasing hormone and adrenocorticotropic hormone and leads to low levels of cortisol, DHEA, DHEAS, and androstenedione.[24] In addition, pharmacological doses of glucocorticoids in amounts greater than 10 mg of prednisone daily or equivalent doses of other glucocorticoids directly inhibit ovarian steroidogenesis.[25] Human immunodeficiency virus (HIV) infection and chronic illness are examples of systemic causes of androgen deficiency.

TESTOSTERONE AND SEXUAL FUNCTION

Table 2 summarizes the effects of testosterone administration on sexual function. The prevalent dogma is that androgens regulate libido in women, although a woman's sexual behavior is greatly affected by environmental, emotional, cultural, and hormonal factors.[21] The effects of androgens in the brain are mediated directly through the androgen receptor and through aromatization of testosterone to estradiol. Androgen receptors have been identified in the cortex, pituitary, hypo-

thalamus, preoptic region, thalamus, amygdala, and brainstem.[26] Testosterone supplementation is associated with increased well-being, energy, appetite, and improved somatic and psychological scores in surgically menopausal women.[27] For instance, in one study of surgically menopausal women, supraphysiological doses of testosterone enanthate alone or in combination with estrogen increased sexual desire, fantasies, and arousal more than estrogen alone.[28] In another study, testosterone and estradiol implants increased sexual activity, satisfaction, pleasure, and frequency of orgasm more than estrogen implants alone.[29]

In a recent, well-designed, placebo-controlled randomized clinical trial, women who underwent hysterectomy and ovariectomy and were on estrogen replacement were randomized to placebo patches or testosterone patches designed to nominally deliver 150 and 300 μg of testosterone daily for 12 weeks each. Although both dose-regimens of testosterone significantly increased serum testosterone levels, only the higher dose, which increased mean serum free testosterone levels into the upper end of the normal female range, was associated with improvements in frequency of sexual activity, pleasure, orgasm, sexual fantasies, masturbation, and positive well-being.[30] Tutten et al. reported that oral administration of testosterone undecenoate increased vaginal vasocongestion as measured by vaginal plethysmography during exposure to a potent visual stimulus in a small number of women with hypothalamic amenorrhea.[29]

In a placebo-controlled crossover study, daily administration of 50 mg DHEA daily for 4 months in women with adrenal insufficiency improved several aspects of sexual function and sense of well-being. It is unclear whether these effects were direct effects of DHEA on the brain or indirect effects due to the conversion of DHEA to testosterone.[31] In contrast, a cross-sectional study did not show a correlation between sexual function and gonadal steroids.[32]

Thus, it appears likely that supraphysiological doses of testosterone that increase serum testosterone levels above the physiological range for healthy younger women may improve some aspects of sexual function in a subset of women with low androgen levels, but we do not know whether physiological replacement doses that increase serum testosterone levels into the mid-range for younger menstruating women would produce meaningful improvements in sexual function and activity in healthy older women with low testosterone levels.

EFFECTS OF TESTOSTERONE ADMINISTRATION ON BODY COMPOSITION, MUSCLE PERFORMANCE, AND PHYSICAL FUNCTION

There is agreement that testosterone administration to men is associated with a dose- and concentration-dependent increase in fat-free mass, muscle size, and maximal voluntary strength and a decrease in fat mass.[33–38] The data on the effects of testosterone administration in women are far more limited.

Table 2. The Effects of Testosterone Administration on Sexual Function

Study	Androgen Formulation and Dose	Duration	Study Design	Patient Group	Effects
Myers, 1990[32]	Conjugated equine estrogen 0.625 mg daily plus methyltestosterone 5 mg daily	4 weeks	Double-blind, placebo-controlled	Naturally menopausal women	Increased pleasure from masturbation, but no improvements in other components of sexual function
Arlt, 2000[31]	DHEA 50 mg daily	4 months	Double-blind, placebo-controlled, cross-over	Women with adrenal insufficiency	Improvements in sexual function and well-being
Sherwin, 1985[28]	Testosterone enanthate 150 mg IM every 4 weeks	Monthly injection for 2 months	Prospective placebo-controlled cross-over	Surgical menopause	Increased sexual fantasies, arousal, desire, somatic and psychological scores
Davis 1995[42]	Testosterone implants 50 mg plus estradiol implants 50 mg versus estradiol implants 50 mg alone	Every 3 months for 2 years	Prospective, single-blind	Postmenopausal women	Increased sexual fantasies, orgasm, and several other aspects of sexual function
Shifren, 2000[30]	Conjugated equine estrogen 0.625 mg, transdermal testosterone 150 μg daily or 300 μg daily	12 weeks	Double-blind, placebo-controlled	Surgically menopausal women	Increased sexual activity, pleasure, orgasm, increased sexual fantasies, masturbation, and positive well-being with 300 μg dose
Tutten 1996[29]	Testosterone undecenoate 40 mg daily	8 weeks	Double-blind, placebo-controlled	Women with amenorrhea	Increased vaginal vasocongestion during exposure to potent visual stimulus

DHEA = dehydroepiandrosterone.
IM = intramuscular.

Total lean mass and lean leg mass are significantly correlated with free but not total testosterone levels in postmenopausal women aged 46 to 55.[39] One cross-sectional study showed that testosterone level predicted muscle strength in postmenopausal women.[40] Estrogen therapy in postmenopausal women, by lowering free testosterone concentrations, can accelerate the loss of lean body mass.[39]

The data on the anabolic effects of testosterone in women are limited. Kenyon et al. reported significant nitrogen retention with administration of pharmacological doses of testosterone propionate to healthy menstruating women.[41] In a more recent study, combined administration of testosterone and estrogen implants increased lean body mass and decreased body fat more than estrogen implants alone.[42] In a placebo-controlled study of HIV-infected women with weight loss, testosterone supplementation by means of transdermal testosterone patches, designed to nominally deliver 150 or 300 μg of testosterone daily, was not associated with significant gains in lean body mass or muscle strength at either testosterone dose (Table 3).[43]

DHEA administration has been reported to increase fat-free mass in postmenopausal women,[44,45] but, in another study, DHEA (50 mg/day) given for 4 months to women with adrenal insufficiency did not result in a change in body mass index or waist-hip ratio (Table 3).[46]

Most of the published studies that have examined the effects of androgen supplementation in women have had small sample sizes, few examined the effects on muscle performance and physical function, and none has unequivocally demonstrated improvements in health-related outcomes. Therefore, it remains unclear whether increasing testosterone concentrations in older women with low testosterone levels into the mid- to upper-normal range will be associated with clinically significant gains in fat-free mass, muscle performance or physical function.

TESTOSTERONE AND COGNITIVE FUNCTION

Most of the research on the hormonal contribution to cognitive function has focused on estrogens. The neuroprotective and neurotrophic effects of estrogens are well recognized, but the underlying mechanisms are not well understood. Estrogens interact with nerve growth factor, brain derived neurotrophic factor, insulin-like growth factor-1 (IGF-1), and fibroblast growth factor.[47–51] Estrogens also act directly on neurotransmitter complexes and ion channels, exhibit antioxidant effects in the brain, reduce neuronal death after exposure to pro-oxidants, and improve cerebral blood flow.[52–54]

Testosterone is aromatized to estradiol in the brain, and some effects of testosterone may be mediated through its conversion to estradiol, but androgen receptors are expressed in specific regions of the brain[55] and likely mediate some of testosterone's organizational effects during brain development and some activational effects postnatally.[56,57] There are gender differences in the distribution of androgen receptors in the human hypothalamus.[58] The effects of testosterone on cognitive function conflict. Low DHEAS levels in postmenopausal women do not correlate with cognitive decline.[59] It is possible that brain levels rather than plasma hormone levels are important. In another study in older men, there was a positive correlation between testosterone and bioavailable testosterone levels and global cognitive function and mental control but not visual-spatial skills.[60] An investigation of healthy younger adults showed that salivary testosterone was negatively correlated with visual-spatial and verbal cognitive scores in right-handed men and positively correlated in right-handed women in a curvilinear fashion.[61] This pattern was not evident in left-handed individuals.

The effects of androgens on cognitive function are domain specific. For instance, observations that men outperform women in a variety of visual-spatial skills suggest that androgens enhance visual-spatial skills.[62] Janowsky et al.[63] tested verbal and visual memory, spatial cognition, motor speed, and cognitive flexibility in a group of older men who received 3 months of testosterone supplementation. Testosterone replacement was associated with a significant improvement in spatial cognition only. Serum testosterone levels were not significantly correlated with spatial performance, but estradiol levels showed a significant inverse relationship to spatial performance, suggesting that estradiol inhibits spatial ability. A study of San Bushmen[63] showed that testosterone but not estradiol levels correlate with better spatial ability and worse verbal fluency. Circulating levels of dihydrotestosterone, a metabolite of testosterone, are positively correlated with verbal fluency. Barrett-Conner et al.[64] found an association of total and bioavailable testosterone levels with global cognitive functioning and mental control but not with visual-spatial skills in older women. Other studies[61,65,66] have reported a curvilinear relationship between androgen levels and spatial ability such that women with high testosterone levels and men with low testosterone levels show the best performance. Several small clinical trials of testosterone supplementation and cognition in older hypogonadal men have provided conflicting results. Sih et al.[67] found no effect, whereas other studies[68,69] reported an effect. Hypogonadal men performed worse on tests of verbal fluency than eugonadal men and showed improvement after testosterone replacement.[70] In transsexual men, administration of an anti-androgen and estrogen, before surgery for gender reassignment, decreased anger and aggression, sexual arousability, and spatial skills and increased verbal fluency. Conversely, testosterone administration to women decreased verbal fluency and increased spatial skills.[71,72]

In summary, the reported literature on testosterone and cognition is equivocal, but these inconsistencies should not be interpreted to mean that there is no effect. Prospective randomized placebo-controlled, trials are needed to determine the effects of physiological testosterone replacement on cognitive function in older women.

TESTOSTERONE AND BONE MINERAL DENSITY

Androgens regulate bone mineral density (BMD) and fracture risk via multiple mechanisms. Testosterone inhibits bone resorption through its conversion to estradiol.[73,74] In addition, androgens also directly stimulate cortical osteoblastic bone formation. Androgen receptors have been reported on osteoblasts and osteocytes. In addition to effects that are mediated through the nuclear androgen receptors, androgens may also exert nongenomic effects on osteoblasts. Androgens directly stimulate alkaline phosphatase and type-1-α collagen synthesis by osteoblasts. In addition, they may indirectly regulate osteoblastic activity by

Table 3. The Effects of Testosterone Supplementation on Body Composition and Muscle Function

Study	Androgen Formulation and Dose	Dose/Duration	Study Design	Patient Group	Effects
Davis 1995[42]	Estradiol implants (50 mg) plus testosterone implants (50 mg) versus estradiol implants alone	Every 3 months for 2 years	Single-blind, randomized, placebo-controlled	Postmenopausal women	Increased lean body mass, decreased body fat without change in BMI
Miller 1998[43]	Transdermal testosterone patch 150 μg and 300 μg daily	Twice weekly for 12 weeks	Randomized, placebo-controlled	Women with AIDS wasting	No significant change in lean body mass
Diamond 1996[45]	DHEA 10% cream	Daily for 12 months	Single-blind	Older women 60–70 years old	Increased fat-free mass
Morales 1998[44]	DHEA 100 mg daily	Daily for 6 months	Randomized, double-blind, placebo-controlled, cross-over	Older men and women	Increased total body mass

BMI = body mass index; AIDS = acquired immune deficiency syndrome; DHEA = dehydroepiandrosterone.

modulating the activity of other bone growth regulators such as IGF-1, IGF-2, fibroblast growth factor, and transforming growth factor-α. IGF-1, and insulin-like growth factor binding proteins (IGF-BPs) have important effects on osteoblast proliferation and differentiation. Androgens increase the expression of IGF-1, IGF-BP2, and IGF-BP3 but decrease inhibitory IGF-BP4 in an androgen-responsive human osteoblastic cell line.[75] Testosterone inhibits parathyroid hormone and interleukin-6 (IL-6) activity; these effects might indirectly result in decreased osteoclastogenesis.[76,77]

In men, androgen deficiency is associated with osteoporosis. Androgen replacement in hypogonadal men decreases markers of bone resorption and increases markers of osteoblastic bone formation and cortical bone mass,[78] but the role of androgen deficiency in the pathophysiology of osteoporosis in older women is poorly understood. We do not know whether age-related decline in testosterone contributes to the risk of osteoporosis and fractures in older women. Serum levels of bioavailable testosterone correlate positively with bone mineral density and negatively with N-telopeptide excretion.[79,80] Women with syndromes of androgen excess have higher bone mass than controls.

Raisz et al.[81] compared the effects of conjugated equine estrogen (CEE) alone with a combined regimen of CEE plus 2.5 mg methyl testosterone on markers of bone formation and bone resorption. Compared with CEE alone, methyl testosterone plus CEE produced a greater increase in bone formation markers such as osteocalcin and bone-specific alkaline phosphatase, but the markers of bone resorption, such as hydroxyproline and pyridinoline crosslinks, were not significantly different between the two groups (Table 4).

Davis et al.[42] reported greater increases in BMD in the spine and hip with estrogen plus testosterone implants than with estrogen implants alone. In another study of postmenopausal women, Watts et al.[82] also found that testosterone plus CEE treatment increased BMD, but CEE alone did not. Both of these important studies had small sample sizes and were relatively short. The effects of androgen supplementation on fracture risks in women have not been examined. It is possible that testosterone supplementation might augment muscle mass and quadriceps muscle strength in older women with low testosterone levels. Because quadriceps muscle strength is a major determinant of fall propensity, direct effects of testosterone on the muscle might provide an additional mechanism by which testosterone might reduce fracture risk in older women (Table 4).

Long-term placebo-controlled studies are needed to determine whether testosterone replacement reduces fracture risk in older women. Because the reference of comparison will likely be women receiving HRT, these studies will likely require large sample sizes.

TESTOSTERONE AND IMMUNE FUNCTION

Testosterone regulates several important aspects of immune function. The prevalence of autoimmune diseases is generally higher in women than in men.[83] Decreased levels of androgens were observed in women with systemic lupus erythematosus (SLE), with lower levels correlated to higher disease activity.[83,84] In an animal model of lupus, the development of "lupus-like" syndrome and progression of kidney disease is more rapid in female than male mice.[85,86] In this animal model, castration of males is associated with a more accelerated development of lupus-like syndrome, and testosterone administration to females retards the progression of this syndrome.[87,88] It has been noted that DHEA inhibits IL-6, tumor necrosis factor, and other cytokines by inhibiting nuclear factor-κB, activation of which is associated with worsened disease activity.[89]

Many patients with autoimmune diseases receive glucocorticoids that might cause loss of muscle and bone mass. Theoretically, testosterone administration, in addition to its immunomodulatory effect, might also prevent or reverse the glucocorticoid-induced muscle wasting and osteoporosis in these patients, but the data on the effects of testosterone administration in patients with autoimmune diseases are not clear. Although some trials of androgen administration failed to demonstrate benefits in patients with rheumatoid arthritis or SLE,[90,91] other studies have reported improvements in some intermediate outcomes and disease activity scores.[92–95] In one study, DHEA administration facilitated withdrawal from glucocorticoid

Table 4. Effect of Testosterone on Bone Mineral Density

Study	Androgen Formulation and Dose	Duration	Study Design	Patient Group	Effects
Raisz, 1996[81]	Conjugated equine estrogen 1.25 mg plus methyl testosterone 2.5 mg daily versus conjugated equine estrogen alone	9 weeks	Double-blind, randomized	Postmenopausal women	Greater increase in bone formation markers (osteocalcin and bone-specific alkaline phosphatase) with combined treatment than with estrogen alone
Davis, 1995[42]	Testosterone implants 50 mg plus estradiol implants versus estradiol implants alone	Every 3 months for 2 years	Single-blind, randomized, placebo-controlled	Postmenopausal women	Greater increase in bone mineral density in the spine and hip in combined treatment group than with estradiol alone
Watts, 1995[82]	Conjugated equine estrogen 1.25 mg plus methyl testosterone 2.5 mg daily versus conjugated equine estrogen alone	Once daily for 2 years	Double-blind, randomized, parallel group	Postmenopausal women	Greater increase in bone mineral density with combined treatment than with estrogen alone

therapy in patients with SLE.[94] Whether DHEA can ameliorate the deleterious effects of glucocorticoids on muscles, bones, and endothelium remains to be determined in randomized clinical trials.[96]

ADVERSE EFFECTS ASSOCIATED WITH ANDROGEN REPLACEMENT

The potential deleterious effects of androgen supplementation in women include the risks of virilization, hirsutism, acne, voice change, erythrocytosis, alterations in plasma lipids and apolipoproteins, and liver toxicity.[97,98] Abnormalities of liver enzymes have been reported with orally administered, 17-α alkylated androgens. Hirsutism is uncommon if supraphysiological levels are avoided.[42,81,99–101] There is concern regarding the increase in cardiovascular risk through lowering of high-density lipoprotein (HDL) levels in women receiving long-term androgen therapy. Davis et al.[42] found no significant change in HDL cholesterol levels in women treated with combined estrogen and testosterone implants, although there was a reduction in total cholesterol and low-density lipoprotein (LDL) cholesterol levels. In another study, combined administration of estrogen and 17-α-methyl testosterone resulted in a 25% reduction in total cholesterol and a modest decrease in HDL2 and HDL3 levels.[81] Some[102,103] but not all studies[44,104] have shown DHEA treatment in women to have negative effects on lipids and lipoproteins. It is unclear whether these adverse effects on plasma lipid profile are limited to supraphysiologic doses of androgens or whether physiologic; testosterone replacement can be administered without significant adverse effects on cardiovascular risk factors.

Hyperandrogenemia is associated with insulin resistance in adolescent girls and women with polycystic ovary syndrome (PCOS).[105,106] Endothelial dysfunction, associated with elevated levels of androgens, may lead to increased risk of macrovascular complications,[105] but consistent correlation between insulin resistance and hyperandrogenemia has not been found in all studies.[106,107] It is unclear whether insulin resistance in patients with PCOS is due to high androgen levels or is inherited as an independent trait. It has been speculated that androgen effects on insulin sensitivity might be biphasic. Holmang et al.[108] demonstrated that, in a rat model, physiological doses of testosterone improve insulin sensitivity, whereas higher doses induce insulin resistance.[109,110]

A recent epidemiological study found an inverse correlation between serum testosterone concentrations and carotid intima-media thickness, a measure of generalized atherosclerosis.[111,112] In LDL-receptor-deficient mice, testosterone administration retards the development of early atherosclerotic lesions. Therefore, it remains unclear whether physiological testosterone replacement in older women will increase or decrease the risk of atherosclerotic heart disease.

METHODS OF ANDROGEN ADMINISTRATION

An oral formulation of testosterone, 17-α-methyltestosterone, approved for use in women in the United States, is typically administered orally in combination with estrogens in doses between 1.25 and 2.5 mg daily. It may be associated with liver toxicity.[97,98]

Other formulations that have been empirically used in women include testosterone esters, testosterone pellets, and testosterone undecenoate given orally in oleic acid (not available in the United States). Testosterone esters, enanthate and cypionate, and the androgenic steroid, nandrolone decanoate, are used traditionally at doses of 25 to 50 mg every 4 weeks, but this regimen may lead to supraphysiological androgen concentrations in the first few days after the injection and suboptimal levels during the last 10 days of the dosing interval. Testosterone implants, administered at a dose of 50 mg every 4 to 6 months, have been used widely outside the United States, but, because of the need for skin incision and insertion through a trocar and the small incidence of spontaneous extrusion, the implants have not been popular in the United States. Oral micronized DHEA at a dose of 25 to 50 mg daily has also been used in clinical trials, but there is considerable batch-to-batch and brand-to-brand variability in the amount of DHEA in various formulations sold over the counter. In addition, the absorption of DHEA from the gastrointestinal tract is variable, and its efficacy in postmenopausal women in improving health-related outcomes has not been established. It should be emphasized that only limited amounts of pharmacokinetic data are available

about these formulations in women, and the regimens used in clinical practice have generally not been physiological.

Two novel testosterone formulations, specifically for use in women, are currently in development. A transdermal testosterone matrix patch has been designed for twice weekly application to the skin. Each transdermal testosterone matrix system (TMTDS) nominally delivers 150 μg of testosterone per day. Thus, two patches applied twice a week can deliver 300 μg of testosterone daily, approximating the daily production rates of testosterone in healthy menstruating women. The pharmacokinetics of transdermally administered testosterone have been studied in premenopausal women, surgically menopausal women, and HIV-infected women.[30,113] These studies have demonstrated that a regimen of two TMTDS patches applied twice a week can maintain serum total and free testosterone levels in the upper-normal range, respectively, in pre- and postmenopausal women with low testosterone levels. Application of each TMTDS patch increases serum total testosterone concentrations on average by 20 to 25 ng/dL.[113] The increments in serum total and free testosterone levels are lower in HIV-infected women treated with TMTDS than healthy women, presumably because of increased plasma clearance or decreased absorption. The skin tolerability of the TMTDS patch has been excellent, with only a small proportion of treated women experiencing mild erythema at the patch application site.

A 1% hydro-alcoholic testosterone gel is under development for use in women; its potential advantages include ease of delivery, invisibility after application, and good skin tolerability. We postulate that testosterone patches and gels will be the most widely used forms of androgen replacement for women in the future, leading to high patient satisfaction and the ability to tolerate dosing to achieve testosterone concentrations in the upper-normal range.

CONCLUSION

Because of the limited availability of sensitive and accurate assays for the measurement of total and free testosterone levels in women and the paucity of normative data, it has been difficult to biochemically define androgen deficiency in women. Although there is tremendous interest in exploring the clinical applications of testosterone supplementation in older women for a number of clinical indications, we do not know whether physiological testosterone replacement can improve clinically relevant outcomes such as physical function, muscle mass and performance, fracture risk, cognitive function, and sexual function. The long-term risks of virilizing side effects and cardiovascular disease also remain largely unknown. Therefore, it would be premature to make a general recommendation about testosterone replacement for all older women with low testosterone levels. At this time, testosterone supplementation in women with the symptom complex that has been loosely named "female androgen deficiency syndrome" should be individualized and preceded by a discussion about the uncertainty of beneficial effects and the potential risks of long-term androgen administration. Further studies are needed to establish testosterone dose-response relationships in women to determine whether it is possible to dissociate the clinically beneficial effects from potential adverse effects.

REFERENCES

1. Sinha-Hikim I, Arver S, Beall G et al. The use of a sensitive equilibrium dialysis method for the measurement of free testosterone levels in healthy, cycling women and in human immunodeficiency virus-infected women. J Clin Endocrinol Metab 1998;83:1312–1318.
2. Southren AL, Gordon GG, Tochimoto S. Further study of factors affecting the metabolic clearance rate of testosterone in man. J Clin Endocrinol Metab 1968;28:1105–1112.
3. Abraham D, Carpenter PC. Issues concerning androgen replacement therapy in postmenopausal women. Mayo Clin Proc 1997;72:1051–1055.
4. Vermeulen A, Ando S. Prolactin and adrenal androgen secretion. Clin Endocrinol (Oxf) 1978;8:295–303.
5. Judd HL, Yen SS. Serum androstenedione and testosterone levels during the menstrual cycle. J Clin Endocrinol Metab 1973;36:475–481.
6. Judd HL, Lucas WE, Yen SS. Effect of oophorectomy on circulating testosterone and androstenedione levels in patients with endometrial cancer. Am J Obstet Gynecol 1974;118:793–798.
7. Chakravarti S, Collins WP, Forecast JD et al. Hormonal profiles after the menopause. BMJ 1976;2:784–787.
8. Zumoff B, Rosenfeld RS, Strain GW et al. Sex differences in the twenty-four-hour mean plasma concentrations of dehydroisoandrosterone (DHA) and dehydroisoandrosterone sulfate (DHAS) and the DHA to DHAS ratio in normal adults. J Clin Endocrinol Metab 1980;51:330–333.
9. Adashi EY. The climacteric ovary as a functional gonadotropin-driven androgen-producing gland. Fertil Steril 1994;62:20–27.
10. Burger HG, Dudley EC, Cui J et al. A prospective longitudinal study of serum testosterone, dehydroepiandrosterone sulfate, and sex hormone-binding globulin levels through the menopause transition. J Clin Endocrinol Metab 2000;85:2832–2838.
11. Sherwin BB, Gelfand MM. The role of androgen in the maintenance of sexual functioning in oophorectomized women. Psychosom Med 1987;49:397–409.
12. Chen W, Zouboulis CC, Fritsch M et al. Evidence of heterogeneity and quantitative differences of the type 1 5alpha-reductase expression in cultured human skin cells—evidence of its presence in melanocytes. J Invest Dermatol 1998;110:84–89.
13. Lombardi G, Zarrilli S, Colao A et al. Estrogens and health in males. Mol Cell Endocrinol 2001;178:51–55.
14. Labrie F, Belanger A, Cusan L et al. Marked decline in serum concentrations of adrenal C19 sex steroid precursors and conjugated androgen metabolites during aging. J Clin Endocrinol Metab 1997;82:2396–2402.
15. Labrie F. Intracrinology. Mol Cell Endocrinol 1991;78:C113–C118.
16. Labrie F, Dupont A, Simard J et al. Intracrinology: The basis for the rational design of endocrine therapy at all stages of prostate cancer. Eur Urol 1993;24:94–105.
17. Dunn JF, Nisula BC, Rodbard D. Transport of steroid hormones: Binding of 21 endogenous steroids to both testosterone-binding globulin and corticosteroid-binding globulin in human plasma. J Clin Endocrinol Metab 1981;53:58–68.
18. Pugeat M, Crave JC, Tourniaire J et al. Clinical utility of sex hormone-binding globulin measurement. Horm Res 1996;45:148–155.
19. Vermeulen A, Verdonck L, Kaufman JM. A critical evaluation of simple methods for the estimation of free testosterone in serum. J Clin Endocrinol Metab 1999;84:3666–3672.
20. Rosner W. An extraordinarily inaccurate assay for free testosterone is still with us. J Clin Endocrinol Metab 2001;86:2903.
21. Sands R, Studd J. Exogenous androgens in postmenopausal women. Am J Med 1995;98:76S–79S.
22. Hojbjerg Gravholt C, Svenstrup B, Bennett P et al. Reduced androgen levels in adult turner syndrome: Influence of female sex steroids and growth hormone status. Clin Endocrinol (Oxf) 1999;50:791–800.
23. Miller KK, Sesmilo G, Schiller A et al. Androgen deficiency in women with hypopituitarism. J Clin Endocrinol Metab 2001;86:561–567.
24. Arlt W, Justl HG, Callies F et al. Oral dehydroepiandrosterone for adrenal androgen replacement: Pharmacokinetics and peripheral conversion to androgens and estrogens in young healthy females after dexamethasone suppression. J Clin Endocrinol Metab 1998;83:1928–1934.
25. Wajchenberg BL, Achando SS, Okada H et al. Determination of the source(s) of androgen overproduction in hirsutism associated with polycystic ovary syndrome by simultaneous adrenal and ovarian venous catheterization. Comparison with the dexamethasone suppression test. J Clin Endocrinol Metab 1986;63:1204–1210.
26. Sarrel PM. Psychosexual effects of menopause: Role of androgens. Am J Obstet Gynecol 1999;180:S319–S324.
27. Sherwin BB, Gelfand MM. Differential symptom response to parenteral estrogen and/or androgen administration in the surgical menopause. Am J Obstet Gynecol 1985;151:153–160.

28. Sherwin BB, Gelfand MM, Brender W. Androgen enhances sexual motivation in females: A prospective, crossover study of sex steroid administration in the surgical menopause. Psychosom Med 1985;47:339–351.
29. Tutten A, Laan E, Panhuysen G et al. Discrepancies between genital responses and subjective sexual function during testosterone substitution in women with hypothalamic amenorrhea. Psychosom Med 1996;58:234–241.
30. Shifren JL, Braunstein GD, Simon JA et al. Transdermal testosterone treatment in women with impaired sexual function after oophorectomy. N Engl J Med 2000;343:682–688.
31. Arlt W, Callies F, Allolio B. DHEA replacement in women with adrenal insufficiency—pharmacokinetics, bioconversion and clinical effects on well-being, sexuality and cognition. Endocr Res 2000;26:505–511.
32. Myers LS, Dixen J, Morrissette D et al. Effects of estrogen, androgen, and progestin on sexual psychophysiology and behavior in postmenopausal women. J Clin Endocrinol Metab 1990;70:1124–1131.
33. Wang C, Swedloff RS, Iranmanesh A et al. Transdermal testosterone gel improves sexual function, mood, muscle strength, and body composition parameters in hypogonadal men. Testosterone Gel Study Group. J Clin Endocrinol Metab 2000;85:2839–2853.
34. Katznelson L, Finkelstein JS, Schoenfeld DA et al. Increase in bone density and lean body mass during testosterone administration in men with acquired hypogonadism. J Clin Endocrinol Metab 1996;81:4358–4365.
35. Brodsky IG, Balagopal P, Nair KS. Effects of testosterone replacement on muscle mass and muscle protein synthesis in hypogonadal men—a clinical research center study. J Clin Endocrin Metab 1996;81:3469–3475.
36. Bhasin S, Storer TW, Berman N et al. Testosterone replacement increases fat-free mass and muscle size in hypogonadal men. J Clin Endocrinol Metab 1997; 82:407–413.
37. Bhasin S, Woodhouse L, Storer T. Proof of the effect of testosterone on skeletal muscle. J Endocrinol 2001;170:27–38.
38. Bhasin S. The dose-dependent effects of testosterone on sexual function and on muscle mass and function. Mayo Clin Proc 2000;75:S70–S76.
39. Gower BA, Nyman L. Associations among oral estrogen use, free testosterone concentration, and lean body mass among postmenopausal women. J Clin Endocrinol Metab 2000;85:4476–4480.
40. Hakkinen K, Pakarinen A. Muscle strength and serum testosterone, cortisol and SHBG concentrations in middle-aged and elderly men and women. Acta Physiol Scand 1993;148:199–207.
41. Kenyon AT, Sandiford I, Bryan AM et al. The effect of testosterone propionate on nitrogen, electrolyte, water and energy metabolism in eunuchoidism. Endocrinology 1938;23:135–144.
42. Davis SR, McCloud P, Strauss BJ et al. Testosterone enhances estradiol's effects on postmenopausal bone density and sexuality. Maturitas 1995;21: 227–236.
43. Miller K, Corcoran C, Armstrong C et al. Transdermal testosterone administration in women with acquired immunodeficiency syndrome wasting: A pilot study. J Clin Endocrinol Metab 1998;83:2717–2725.
44. Morales AJ, Haubrich RH, Hwang JY et al. The effect of six months treatment with a 100 mg daily dose of dehydroepiandrosterone (DHEA) on circulating sex steroids, body composition and muscle strength in age-advanced men and women. Clin Endocrinol (Oxf) 1998;49:421–432.
45. Diamond P, Cusan L, Gomez JL et al. Metabolic effects of 12-month percutaneous dehydroepiandrosterone replacement therapy in postmenopausal women. J Endocrinol 1996;150:S43–S50.
46. Callies F, Fassnacht M, van Vlijmen JC et al. Dehydroepiandrosterone replacement in women with adrenal insufficiency: Effects on body composition, serum leptin, bone turnover, and exercise capacity. J Clin Endocrinol Metab 2001;86:1968–1972.
47. Fernandez-Galaz MC, Morschl E, Chowen JA et al. Role of astroglia and insulin-like growth factor-I in gonadal hormone-dependent synaptic plasticity. Brain Res Bull 1997;44:525–531.
48. Gibbs RB, Wu D, Hersh LB et al. Effects of estrogen replacement on the relative levels of choline acetyltransferase, trkA, and nerve growth factor messenger RNAs in the basal forebrain and hippocampal formation of adult rats. Exp Neurol 1994;129:70–80.
49. Murphy DD, Cole NB, Segal M. Brain-derived neurotrophic factor mediates estradiol-induced dendritic spine formation in hippocampal neurons. Proc Natl Acad Sci U S A 1998;95:11412–11417.
50. Takahashi H, Nakagawa S. Effects of estrogen on cell growth and fibroblast growth factor receptor induction in MtT/Se cells. Endocr Res 1997; 23:95–104.
51. Toran-Allerand CD. Mechanisms of estrogen action during neural development: Mediation by interactions with the neurotrophins and their receptors? J Steroid Biochem Mol Biol 1996;56:169–178.
52. Singer CA, Rogers KL, Dorsa DM. Modulation of Bcl-2 expression: A potential component of estrogen protection in NT2 neurons. Neuroreport 1998;9:2565–2568.
53. Ross RL, Gu Q, Wong M. Estrogen: Nontranscriptional signaling pathway. Rec Prog Horm Res 1997;52:33–68.
54. Goodman Y, Bruce AJ, Cheng B et al. Estrogens attenuate and corticosterone exacerbates excitotoxicity, oxidative injury, and amyloid beta-peptide toxicity in hippocampal neurons. J Neurochem 1996;66:1836–1844.
55. Simerly RB, Chang C, Muramatsu M et al. Distribution of androgen and estrogen receptor mRNA-containing cells in the rat brain: An in situ hybridization study. J Comp Neurol 1990;294:76–95.
56. Lu S, Simon NG, Wang Y et al. Neural androgen receptor regulation: Effects of androgen and antiandrogen. J Neurobiol 1999;41:505–512.
57. Dorner G, Hinz G. Androgen dependent brain differentiation and life span. Endokrinologie 1975;65:378–380.
58. Fernandez-Guasti A, Kruijver FP, Fodor M et al. Sex differences in the distribution of androgen receptors in the human hypothalmus. J Comp Neurol 2000;425:422–435.
59. Barrett-Connor E, Edelstein SL. A prospective study of dehydrepiandrosterone sulfate and cognitive function in an older population: The Rancho Bernardo Study. J Am Geriatr Soc 1994;42:420–423.
60. Barrett-Connor E, Goodman-Gruen D, Patay B. Endogenous sex hormones and cognitive function in older men. J Clin Endocrinol Metab 1999;84: 3681–3685.
61. Moffat SD, Hampson E A. curvilinear relationship between testosterone and spatial cognition in humans: Possible influence of hand preference. Psychoneuroendocrinology 1996;21:323–337.
62. Christiansen K. Sex hormone-related variations of cognitive performance in Kung San hunter-gatherers of Namibia. Neuropsychobiology 1993;27:97–107.
63. Janowsky JS, Oviatt SK, Orwoll ES. Testosterone influences spatial cognition in older men. Behav Neurosci 1994;108:325–332.
64. Barrett-Connor E, Goodman-Gruen D. Cognitive function and endogenous sex hormones in older women. J Am Geriatr Soc 1999;47:1289–1293.
65. Gouchie C, Kimura D. The relationship between testosterone levels and cognitive ability patterns. Psychoneuroendocrinology 1991;16:323–334.
66. Shute VJ PJ, Hubert L, Reynolds RW. The relationship between androgen levels and human spatial abilities. Bull Psychonomic Soc 1983;21:465–468.
67. Sih R, Morley JE, Kaiser FE et al. Testosterone replacement in older hypogonadal men: A 12-month randomized controlled trial. J Clin Endocrinol Metab 1997;82:1661–1667.
68. Cherrier MM, Asthana S, Plymate S et al. Testosterone supplementation improves spatial and verbal memory in healthy older men. Neurology 2001;57:80–88.
69. Herbst KL, Anawalt BD, Cherrie RM et al. Testosterone administration improves spatial and verbal memory in normal men. J Invest Med 1999; 47:23A.
70. Alexander GM, Swerdloff RS, Wang C et al. Androgen-behavior correlations in hypogonadal men and eugonadal men. II. Cognitive abilities. Horm Behav 1998;33:85–94.
71. Van Goozen SH, Cohen-Kettenis PT, Gooren LJ et al. Gender differences in behaviour: Activating effects of cross-sex hormones. Psychoneuroendocrinology 1995;20:343–363.
72. Slabbekoorn D, van Goozen SH, Megens J et al. Activating effects of cross-sex hormones on cognitive functioning: A study of short-term and long-term hormone effects in transsexuals. Psychoneuroendocrinology 1999;24:423–447.
73. Khosla S, Melton LJ, III, Atkinson EJ et al. Relationship of serum sex steroid levels and bone turnover markers with bone mineral density in men and women: A key role for bioavailable estrogen. J Clin Endocrinol Metab 1998; 83:2266–2274.
74. Falahati-Nini A, Riggs BL, Atkinson EJ et al. Relative contributions of testosterone and estrogen in regulating bone resorption and formation in normal elderly men. J Clin Invest 2000;106:1553–1560.
75. Gori F, Hofbauer LC, Conover CA et al. Effects of androgens on the insulin-like growth factor system in an androgen-responsive human osteoblastic cell line. Endocrinology 1999;140:5579–5586.
76. Ershler WB, Keller ET. Age-associated increased interleukin-6 gene expression, late-life diseases, and frailty. Annu Rev Med 2000;51:245–270.
77. Chen Q, Kaji H, Sugimoto T et al. Testosterone inhibits osteoclast formation stimulated by parathyroid hormone through androgen receptor. FEBS Lett 2001;491:91–93.
78. Kamel HK, Perry HM III, Morley JE. Hormone replacement therapy and fractures in older adults. J Am Geriatr Soc 2001;49:179–187.
79. Shoupe D. Androgens and bone: Clinical implications for menopausal women. Am J Obstet Gynecol 1999;180:S329–S333.
80. Zofkova I, Bahbouh R, Hill M. The pathophysiological implications of circulating androgens on bone mineral density in a normal female population. Steroids 2000;65:857–861.
81. Raisz LG, Wiita B, Artis A et al. Comparison of the effects of estrogen alone and estrogen plus androgen on biochemical markers of bone formation and resorption in postmenopausal women. J Clin Endocrinol Metab 1996;81:37–43.

82. Watts NB, Notelovitz M, Timmons MC et al. Comparison of oral estrogens and estrogens plus androgen on bone mineral density, menopausal symptoms, and lipid-lipoprotein profiles in surgical menopause. Obstet Gynecol 1995;85:529–537.
83. Lahita RG, Bradlow HL, Ginzler E et al. Low plasma androgens in women with systemic lupus erythematosus. Arthritis Rheum 1987;30:241–248.
84. Jungers P, Nahoul K, Pelissier C et al. Plasma androgens in women with disseminated lupus erythematosus. Presse Med 1983;12:685–688.
85. Dumont F, Monier JC. Sex-dependent systemic lupus erythematosus-like syndrome in (NZB X SJL)F1 mice. Clin Immunol Immunopathol 1983;29:306–317.
86. Carlsten H, Nilsson N, Jonsson R et al. Differential effects of oestrogen in murine lupus: Acceleration of glomerulonephritis and amelioration of T cell-mediated lesions. J Autoimmun 1991;4:845–856.
87. Verheul HA, Deckers GH, Schuurs AH. Effects of nandrolone decanoate in NZB/W mice treated concomitantly with maintenance doses of dexamethasone sodium phosphate. Int J Immunopharmacol 1985;7:249–254.
88. Verheul HA, Deckers GH, Schuurs AH. Effects of nandrolone decanoate or testosterone decanoate on murine lupus: Further evidence for a dissociation of autoimmunosuppressive and endocrine effects. Immunopharmacology 1986;11:93–99.
89. Straub RH, Scholmerich J, Zietz B. Replacement therapy with DHEA plus corticosteroids in patients with chronic inflammatory diseases—substitutes of adrenal and sex hormones. Z Rheumatol 2000;59(Suppl 2):II/108–118.
90. Lahita RG, Cheng CY, Monder C et al. Experience with 19-nortestosterone in the therapy of systemic lupus erythematosus: Worsened disease after treatment with 19-nortestosterone in men and lack of improvement in women. J Rheumatol 1992;19:547–555.
91. Hall GM, Larbre JP, Spector TD et al. A randomized trial of testosterone therapy in males with rheumatoid arthritis. Br J Rheumatol 1996;35:568–573.
92. van Vollenhoven RF, Morabito LM, Engleman EG et al. Treatment of systemic lupus erythematosus with dehydroepiandrosterone: 50 patients treated up to 12 months. J Rheumatol 1998;25:285–289.
93. van Vollenhoven RF, Park JL, Genovese MC et al. A double-blind, placebo-controlled, clinical trial of dehydroepiandrosterone in severe systemic lupus erythematosus. Lupus 1999;8:181–187.
94. van Vollenhoven RF. Dehydroepiandrosterone in systemic lupus erythematosus. Rheum Dis Clin North Am 2000;26:349–362.
95. Isenberg D, Ramsey-Goldman R. Assessing patients with lupus: Towards a drug responder index. Rheumatology (Oxford) 1999;38:1045–1049.
96. Robinzon B, Cutolo M. Should dehydroepiandrosterone replacement therapy be provided with glucocorticoids? Rheumatology (Oxford) 1999;38:488–495.
97. Lobo RA. Androgens in postmenopausal women: Production, possible role, and replacement options. Obstet Gynecol Surv 2001;56:361–376.
98. Westaby D, Ogle SJ, Paradinas FJ et al. Liver damage from long-term methyltestosterone. Lancet 1977;2:262–263.
99. Davis SR, Burger HG. Clinical review 82: Androgens and the postmenopausal woman. J Clin Endocrinol Metab 1996;81:2759–2763.
100. Burger HG, Hailes J, Menelaus M et al. The management of persistent menopausal symptoms with oestradiol-testosterone implants: Clinical, lipid and hormonal results. Maturitas 1984;6:351–358.
101. Burger H, Hailes J, Nelson J et al. Effect of combined implants of oestradiol and testosterone on libido in postmenopausal women. BMJ (Clin Res Ed) 1987;294:936–937.
102. Mortola JF, Yen SS. The effects of oral dehydroepiandrosterone on endocrine-metabolic parameters in postmenopausal women. J Clin Endocrinol Metab 1990;71:696–704.
103. Casson PR, Santoro N, Elkind-Hirsch K et al. Postmenopausal dehydroepiandrosterone administration increases free insulin-like growth factor-I and decreases high-density lipoprotein: A six-month trial. Fertil Steril 1998;70:107–110.
104. Barnhart KT, Freeman E, Grisso JA et al. The effect of dehydroepiandrosterone supplementation to symptomatic perimenopausal women on serum endocrine profiles, lipid parameters, and health-related quality of life. J Clin Endocrinol Metab 1999;84:3896–3902.
105. Paradisi G, Steinberg HO, Hempfling A et al. Polycystic ovary syndrome is associated with endothelial dysfunction. Circulation 2001;103:1410–1415.
106. Scarpitta AM, Sinagra D. Polycystic ovary syndrome: An endocrine and metabolic disease. Gynecol Endocrinol 2000;14:392–395.
107. Mather KJ, Verma S, Corenblum B et al. Normal endothelial function despite insulin resistance in healthy women with the polycystic ovary syndrome. J Clin Endocrinol Metab 2000;85:1851–1856.
108. Holmang A, Bjorntorp P. The effects of testosterone on insulin sensitivity in male rats. Acta Physiol Scand 1992;146:505–510.
109. Rizza RA. Androgen effect on insulin action and glucose metabolism. Mayo Clin Proc 2000;75(Suppl):S61–S64.
110. Usiskin KS, Butterworth S, Clore JN et al. Lack of effect of dehydroepiandrosterone in obese men. Int J Obes 1990;14:457–463.
111. Bernini GP, Moretti A, Sgro M et al. Influence of endogenous androgens on carotid wall in postmenopausal women. Menopause 2001;8:43–50.
112. Bernini GP, Sgro M, Moretti A et al. Endogenous androgens and carotid intimal-medial thickness in women. J Clin Endocrinol Metab 1999;84:2008–2012.
113. Javanbakht M, Singh AB, Mazer NA et al. Pharmacokinetics of a novel testosterone matrix transdermal system in healthy, premenopausal women and women infected with the human immunodeficiency virus. J Clin Endocrinol Metab 2000;85:2395–2401.