CLINICAL STUDY

Influence of methyltestosterone postmenopausal therapy on plasma lipids, inflammatory factors, glucose metabolism and visceral fat: a randomized study

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Abstract

Background: There has been a growing interest in treating postmenopausal women with androgens. However, hyperandrogenemia in females has been associated with increased risk of cardiovascular disease.

Objective: We aimed to assess the effects of androgen replacement on cardiovascular risk factors.

Design: Thirty-seven postmenopausal women aged 42–62 years that had undergone hysterectomy were prospectively enrolled in a double-blind protocol to receive, for 12 months, percutaneous estradiol (E₂) (1 mg/day) combined with either methyltestosterone (MT) (1.25 mg/day) or placebo.

Methods: Along with treatment, we evaluated serum E₂, testosterone, sex hormone-binding globulin (SHBG), free androgen index, lipids, fibrinogen, and C-reactive protein; glucose tolerance; insulin resistance; blood pressure; body-mass index; and visceral and subcutaneous abdominal fat mass as assessed by computed tomography.

Results: A significant reduction in SHBG ($P < 0.001$) and increase in free testosterone index ($P < 0.05$; Repeated measures analysis of variance) were seen in the MT group. Total cholesterol, triglycerides, fibrinogen, and systolic and diastolic blood pressure were significantly lowered to a similar extent by both regimens, but high-density lipoprotein cholesterol decreased only in the androgen group. MT-treated women showed a modest rise in body weight and gained visceral fat mass relative to the other group ($P < 0.05$), but there were no significant detrimental effects on fasting insulin levels and insulin resistance.

Conclusion: This study suggests that the combination of low-dose oral MT and percutaneous E₂, for 1 year, does not result in expressive increase of cardiovascular risk factors. This regimen can be recommended for symptomatic postmenopausal women, although it seems prudent to perform baseline and follow-up lipid profile and assessment of body composition, especially in those at high risk of cardiovascular disease.

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Introduction

Quantitatively, androgens are the predominant sex steroid in women (1). From 30 years of age, women experience a progressive decline in androgen serum levels as a consequence of age-related decrease in adrenal and ovarian production (2–4). This may lead to the clinical syndrome of female androgen insufficiency characterized by insidious tiredness, diminished sense of well-being and libido, alterations in body composition and bone loss. If associated with low free testosterone levels, these signs and symptoms (more severe in the presence of specific etiologic factors, such as bilateral oophorectomy, premature menopause induced by

chemotherapy or irradiation, adrenal insufficiency or adrenal suppression) can usually be relieved by discerning androgen administration, a therapy that has been largely accepted (5, 6).

When currently recommended doses are administered, it appears that the benefits to bone mass, sexuality and general well-being are achieved without significant side effects (7). However, available data do not support the uncontrolled use of androgens in women, because their precise role in atherosclerosis is still unclear and the effects of long-term androgen replacement on cardiovascular disease, the leading cause of death for postmenopausal women in developed countries, are not established.

Restoring physiologic levels of testosterone seems to protect cardiovascular health in postmenopausal women (8, 9), but female hyperandrogenemia, as reflected by low circulating sex hormone-binding globulin (SHBG) concentrations, is associated with reduced high-density lipoprotein (HDL) cholesterol (10, 11), hyperinsulinemia, glucose intolerance, insulin resistance (12) and increased accumulation of abdominal fat (13, 14), all of which contribute to increased risk of cardiovascular disease. It has been proposed that the increased lipolytic activity of visceral fat results in excessive release of free fatty acids into the portal vein (15), resulting in reduced hepatic insulin clearance (16), increased hepatic glucose and very-low-density lipoprotein production (17, 18).

We therefore performed a 1-year, prospective, double-blind, placebo-controlled study to assess the effects of adding low-doses of methyltestosterone (MT) to estrogen replacement on the cardiovascular risk factors of postmenopausal women, including anthropometric data, blood pressure, inflammatory factors, lipid metabolism, glucose metabolism and abdominal fat.

Patients and methods

Fifty hysterectomized volunteers with serum follicle-stimulating hormone (FSH) in the menopausal range (>40 mUI/ml), and younger than 65 years, were recruited from two university medical centers (Fig. 1). A screening evaluation was done, including complete medical history and physical examination, Papanicolaou smear, transvaginal ultrasound scan and mammogram. Subjects were excluded from the study if any of the following conditions were present: acne or hirsutism classified as greater than a Ferriman–Galley score of 8, impaired hepatic or renal function, diabetes mellitus, coronary disease, systolic blood pressure over

180 mmHg or diastolic blood pressure over 110 mmHg, use of any estrogen formulation in the past 3 months and contraindications for estrogen replacement.

The study was approved by the research ethics committee of the hospital of the Federal University of Rio de Janeiro, Brazil. All selected participants gave informed consent upon receiving information regarding the nature, purpose and duration of the study, and were guaranteed continued medical care if they chose to withdraw from the study.

Thirty-seven women, aged 42–62, were prospectively enrolled in a double-blind protocol to receive for 12 months percutaneous estradiol (E_2) (Estreva gel; Merck) 1 mg/day associated with either oral MT 1.25 mg or a placebo provided in identical tablets by a doctor of pharmacy and taken every morning. A simple random allocation was performed by the pharmacist who compounded the tablets.

During the observation period, eligible women returned every 3 months for clinical examination, which was always performed before breakfast. Each patient had her body weight and height recorded while wearing only light clothes and no shoes. Body-mass index (BMI) was calculated as weight divided by height squared (kg/m^2). Waist circumference was measured midway between the lateral lower rib margin and the superior anterior iliac crest while the subjects were standing, and hip circumference was measured similarly at the level of the great trochanters. Waist-to-hip ratio (WHR) was obtained from these two measurements. After subjects rested for at least 15 min, blood pressure was measured in a sitting position, on the right arm with an appropriately sized cuff. Adherence to study medications was assessed by counting tablets in returned bottles.

Blood specimens were collected at 0, 6 and 12 months, between 0800 and 0900 h, after 12-h overnight fast. Hematocrit and white blood cell count, glucose, total

Figure 1 Flowchart showing steps in treatment procedures in this study.

cholesterol, HDL cholesterol, low-density lipoprotein (LDL) cholesterol, triglycerides (TG), urea, creatinine, hepatic transaminases, total bilirubin and total alkaline phosphatase levels were measured by automated methods. A 75 g oral glucose tolerance test (OGTT) was performed at baseline and 12 months. Normal glucose tolerance, impaired glucose tolerance and diabetes were defined according to the Expert Committee on Diagnosis and Classification of Diabetes Mellitus criteria (19). Serum aliquots for determination of total testosterone (T), E₂, SHBG, insulin, C-reactive protein and fibrinogen were kept frozen at -70 °C until analysis, at the end of the study.

T, E₂ and SHBG were analyzed by immunoradiometric assays using commercial kits from Diagnostic Products (DPC, Los Angeles, CA, USA). Direct estimation of serum insulin was performed by fluoroimmunoassay (PerkinElmer, São Paulo, SP, Brazil), C-reactive protein by N high-sensitivity immunonephelometry (Dade Behring, São Paulo, SP, Brazil) and fibrinogen levels by the clotting method of Clauss (Diagnostica Stago, Asnieres, France). The intra-assay coefficients of variation were 7.4% for T, 6.6% for E₂, 5.2% for SHBG, 3.1% for insulin, 2.9% for C-reactive protein and 2.8% for fibrinogen. Reference ranges were 0.347–2.277 nmol/l for T, undetectable 168.86 pmol/l for E₂, 18–114 nmol/l for SHBG, 16.94–191.16 pmol/l (2.34–26.4 µU/ml) for insulin, <0.3 mg/dl for C-reactive protein and 200–400 mg/dl for fibrinogen.

Bioavailable T was assessed by using the free androgen index (FAI) calculated from T and SHBG by the following formula: T nmol/l × 100/SHBG nmol/l (20). Insulin resistance was estimated by the homeostasis model assessment (HOMA) formula: fasting insulin µU/ml × glucose mmol/l / 22.5 (21).

At the beginning and the end of the study, computed tomography (CT) was performed by a single investigator, in the morning, on a Somatom balance scanner (Siemens, Erlanger, Germany) at the imaging division of the university hospital. Technical factors included 120 kVp, variable mA and 9.8-s total scan time. The scan was calibrated to ensure variation of less than -5 Hounsfield units (HU). A single slice (10 mm) was obtained at the level of the umbilicus while patients remained in the supine position, their arms stretched

above the head. Parameters for fat density were selected as the range between -250 and -50 HU by the method of Borkan and Gerzof (22). Each CT slice was analyzed for the cross-sectional area of fat with a density contour program available in the Siemens software. Images were taken for total abdominal adipose tissue volume analysis. To assess intra-abdominal adipose tissue volume, each abdominal image was edited by removing the image exterior to the innermost abdominal wall muscles with a mouse-driven cursor. Replicate measures of intra-abdominal adipose volume revealed a variability of 0.2–0.1 ml per CT slice analyzed.

Statistical analysis was performed with Stata 7 (Stata Corp 2001, www.stata.com). Data were expressed as means ± s.d. or proportions. Baseline characteristics of the two groups were compared by unpaired *t*-test or Fisher's exact test. Analysis of variance for repeated measures (ANOVA) was used to test differences between treatment groups and changes over time. If the treatment × time interaction term was significant (*P* < 0.05) on ANOVA, differences between adjacent values obtained at various time points were compared by paired *t*-tests, with the Bonferroni correction. Data were analyzed by log transformation (SHBG, FAI, fasting insulin, HOMA, HDL cholesterol, LDL cholesterol, and TG) or nonparametric tests (E₂ and C-reactive protein) as appropriate. For the latter variables, differences in variations between 1 year and baseline values were investigated with the Mann–Whitney test. All statistical tests were based on two-tailed alternatives. A *P* value less than 0.05 were accepted as significant for all analysis in the study.

Results

All 37 patients used the study medications regularly for 1 year and were analyzed for outcomes. Sixteen patients received percutaneous E₂ plus MT (MT group) and 21 received percutaneous E₂ plus placebo (placebo group). Only two patients in each group did not have vasomotor symptoms of flushes and sweats.

Table 1 shows some demographic characteristics of the samples. They were well matched for age, race, menopause nature, weight, BMI, WHR, systolic

Table 1 Baseline demographic characteristics of postmenopausal women selected to receive percutaneous estradiol associated with methyltestosterone (MT) or placebo (P).

Variable		MT group (n = 16)	P group (n = 21)	P value
Race: n (%)	White	5 (31.25%)	7 (33.33%)	0.983
	Mulatto	5 (31.25%)	6 (28.55%)	
	Black	6 (37.50%)	8 (38.50%)	
Menopause: n (%)	Natural	8 (50%)	9 (42.86%)	0.666
	Oophorectomy	8 (50%)	12 (57.14%)	
Age (years)		54.06 ± 4.85	52.57 ± 6.26	0.436

Data presented as absolute values (percentages) or means ± s.d. Comparisons using Fisher's exact test or unpaired *t*-test. *P* < 0.05 = statistically significant.

and diastolic blood pressure, metabolic parameters, FAI ($P = 0.60$), E_2 ($P = 0.10$), T ($P = 0.06$), SHBG ($P = 0.52$) and fibrinogen levels. However, mean values of C-reactive protein (0.53–0.49 vs 0.33–0.14 mg/dl, $P = 0.04$) were already higher in the group of women selected to receive MT.

Virilizing side effects, not regarded as serious, were seen in some participants during androgen replacement. There were no complaints of hirsutism, but acne, seborrhea or both were noted in 25% of the subjects, while alopecia affected only one woman. No significant changes in laboratory indices, including hematology values and renal and liver function tests, were seen in either group.

There were no differences between women who had undergone bilateral oophorectomy or not, in baseline serum levels of E_2 (161.89–58 vs 162.99–62.4 pmol/l; $P = 0.948$), T (1.133–0.453 vs 1.253–0.549 nmol/l; $P = 0.195$) and SHBG (51.33–31.65 vs 51.81–36.58 nmol/l; $P = 0.966$) respectively. The variations in T serum levels were also similar in oophorectomized and nonoophorectomized patients, as shown in Table 2. Therefore, they were not assessed separately.

MT and placebo groups were studied for all comparisons. The increases in circulating E_2 levels during the 1-year period were not significantly different in these two groups: 185.6 ± 70.3 to 429.4 ± 305.9 pmol/l in the MT group and 145.1 ± 43 to 490.1 ± 406.2 pmol/l in the placebo group; $P = 0.350$. As shown in Table 2, T did not significantly change with treatment in either the MT group ($P = 0.144$) or the placebo group ($P = 0.441$). Combined treatment produced the expected changes in FAI and SHBG levels. FAI increased ($P < 0.05$) and SHBG decreased ($P < 0.001$) in the MT group throughout the study, as shown in Table 3. Mean values of both parameters, however, remained within normal reference ranges.

Changes in lipid metabolism are also shown in Table 3. Significant reductions in serum cholesterol and TG were seen with both treatments. LDL cholesterol was unchanged, but HDL cholesterol fell from pretreatment levels during the first 6 months, with no further decrease, only in the MT group. Mean HDL cholesterol

was statistically different in the MT and placebo groups at 6 months and when we stopped follow-up.

C-reactive protein levels were unaffected in both groups ($P = 0.35$). Mean values were higher in the MT group at baseline, and this difference persisted after 1 year of therapy (0.56–0.64 mg/dl in the MT group vs 0.39–0.28 mg/dl in the placebo group). On the other hand, a very significant and similar decrease in serum fibrinogen ($P < 0.001$) was documented with both treatments (Table 4).

Statistically significant reductions in systolic blood pressure and diastolic blood pressure were observed in both groups during the study (Table 4).

There was a slight but significant increase in body weight and BMI after androgen use. However, no significant changes to the WHR were noted in either group (Table 4). Within the 1-year study period, both treatments produced similar decreases in total and subcutaneous abdominal fat estimated by CT, although the amount of visceral fat was reduced only in women with isolated percutaneous E_2 . Combined treatment produced a slight increase in intra-abdominal fat mass, causing the between-group difference to become significant (Table 5).

Fasting glucose and 2-h blood glucose levels were not changed by either therapy, but equivalent decreases from baseline values in mean serum insulin and HOMA were observed in both groups at the end of the study (Table 5).

Discussion

Recently, there has been a growing interest in the treatment of postmenopausal women with androgens. Although the addition of low-dose testosterone to estrogen therapy has shown improvements in sexual function (23, 24), body composition (25) and bone mineral density (26) in postmenopausal women, the question remains: what are the effects of this regimen on cardiovascular risk parameters?

Investigations have indicated that coadministration of oral androgens attenuates the cardioprotective effect of

Table 2 Variations in total testosterone (nmol/l) with treatment with percutaneous estradiol associated with methyltestosterone (MT) or placebo (P) in women with natural menopause and bilateral oophorectomy.

	Placebo			Methyltestosterone		
	All <i>n</i> = 21	Nonoophor. <i>n</i> = 9	Oophorect. <i>n</i> = 12	All <i>n</i> = 16	Nonoophor. <i>n</i> = 8	Oophorect. <i>n</i> = 8
Baseline	1.018±0.325	1.042±0.420	1.000±0.251	1.411±0.596	1.490±0.604	1.133±0.619
6 months	0.936±0.339	0.900±0.332	0.964±0.419	1.249±0.488	1.254±0.549	1.244±0.457
12 months	0.935±0.423	0.978±0.468	0.899±0.403	1.204±0.326	1.224±0.300	1.185±0.371
<i>P</i> value						
Time	NS	NS	NS	NS	NS	NS
Interaction	NS	NS	NS	NS	NS	NS

Nonoophor: natural menopause, nonoophorectomized; oophorect: bilateral oophorectomy.

Results (mean±s.d.) of repeated measures ANOVA are reported. Significant differences ($P < 0.05$).

Table 3 Variations in androgen parameters and lipid metabolism with treatment with percutaneous estradiol associated with methyltestosterone (MT, *n* = 16) or placebo (P, *n* = 21) in postmenopausal women.

	FAI	SHBG (nmol/l)	Cholesterol (mg/dl)	HDL cholesterol (mg/dl)	LDL cholesterol (mg/dl)	TG (mg/dl)
MT group						
Baseline	3.7±3.0	53.9±35.3	221.6±37.7	53.9±18.8	150.1±36.3	123.1±61.3
6 months	5.3±4.2	37.2±32.4	198.0±38.9	44.3±11.2	133.2±35.2	101.9±59.7
12 months	5.2±3.2	32.6±23.3	203.3±37.0	44.2±12.9	139.4±34.7	101.7±53.9
P group						
Baseline	3.2±2.3	49.8±32.7	212.7±49.0	53.2±13.9	132.5±44.1	133.9±129.1
6 months	2.4±1.8	54.4±33.4	207.7±41.2	54.3±11.2	135.1±40.4	106.0±45.5
12 months	2.4±1.7	52.2±30.9	207.0±33.8	53.4±13.4	135.6±35.0	96.0±37.0
P value						
Time	NS	<0.001	0.03	NS	NS	0.03
Interaction	0.003	<0.001	NS	0.01	NS	NS

FAI: free androgen index; HDL: high-density lipoprotein; LDL: low-density lipoprotein; SHBG: sex hormone-binding globulin; TG: triglycerides. Results (mean±s.d.) of repeated measures ANOVA are reported. Significant differences.

estrogen on serum HDL; reduces total cholesterol, TG and lipoprotein little a (Lp(a)) (27–29); and has a neutral effect on blood flow (30), while parenteral testosterone therapy does not adversely affect lipoproteins (6) but rather improves both endothelium-dependent and endothelium-independent vasodilation associated with postmenopausal estrogen replacement (31).

In our research, as well as in earlier studies (28, 29, 32), oral MT given in low doses did not interfere with the efficacy of estrogen to reduce cholesterol levels and had no deleterious effects on the LDL fraction of postmenopausal women. We also confirmed the unfavorable decrease in the circulating concentration of HDL cholesterol induced by MT (28, 29). Significant reductions were noted at 6 and 12 months of therapy, and in three MT users who had low pretreatment HDL cholesterol values, HDL cholesterol levels were below the desirable range during the observational period.

The clinical significance of these changes with respect to risk factors for cardiovascular disease is unknown, although it has been recently suggested that the androgen-induced HDL decline should not be automatically assumed to be proatherogenic, because

it may instead reflect accelerated reverse cholesterol transport (33). Indeed, epidemiologic and clinical studies provide evidence of the cardioprotective effects of HDL cholesterol level attributed to its role in cholesterol reversal transport but also to its effects on endothelial cells and its antioxidant activity. Further studies are needed to determine the clinical significance of low HDL cholesterol levels (34).

It was previously reported that postmenopausal women treated with combined MT/conjugated estrogen replacement experience a significant reduction in plasma TG when compared with conjugated estrogen-only therapy (29, 35). This finding should be taken into consideration because TG-containing lipoproteins are closely correlated with plasma viscosity (36), an established risk factor for cardiovascular disease (37). Furthermore, there is some evidence in support of TG as an independent risk factor in certain subgroups, including postmenopausal women (38). In our investigation, however, TG concentrations were similar in women who were taking estrogen alone or associated with MT, and this apparent discrepancy is certainly due to the fact that we used percutaneous instead of

Table 4 Variations in fibrinogen, systolic blood pressure (BP), diastolic blood pressure and anthropometric measures with treatment with percutaneous estradiol associated with methyltestosterone (MT, *n* = 16) or placebo (P, *n* = 21) in postmenopausal women.

	Fibrinogen (mg/dl)	Systolic BP (mmHg)	Diastolic BP (mmHg)	Weight (kg)	BMI (kg/m ²)	WHR
MT group						
Baseline	344.5±102.2	127.2±22.6	80.3±14.4	70.6±16.8	28.7±6.8	0.80±0.05
6 months		118.1±20.1	76.9±14.6	72.2±17.6	29.5±6.9	0.78±0.04
12 months	226.5±113.7	121.9±14.2	75.6±10.3	72.8±17.5	29.8±6.9	0.79±0.04
P group						
Baseline	285.3±76.4	129.3±14.0	80.7±9.8	66.0±12.5	27.5±4.6	0.83±0.07
6 months		120.0±12.9	75.7±7.5	66.8±12.7	27.9±4.6	0.83±0.06
12 months	201.6±91.8	124.8±13.6	78.8±8.6	66.5±12.6	27.7±4.7	0.82±0.06
P value						
Time	<0.001	0.002	0.04	0.02	0.01	NS
Interaction	NS	NS	NS	NS	NS	NS

Results (mean±s.d.) of repeated measures ANOVA are reported. Significant differences (*P* < 0.05). BMI: body-mass index; WHR: waist-to-hip ratio.

Table 5 Variations in abdominal fat area and glucose metabolism with treatment with percutaneous estradiol associated with methyltestosterone (MT, $n = 16$ or placebo (P, $n = 21$)) in postmenopausal women.

	Visceral fat area (cm ²)	Subcutaneous fat area (cm ²)	Glucose (mg/dl)	2-h glucose (mg/dl)	Insulin (pmol/l)	HOMA
MT group						
Baseline	103.0±49.4	343.2±148.2	85.8±13.0	108.6±32.8	92.76±50.9	2.7±1.2
6 months			85.8±13.3			
12 months	106.5±59.5	316.1±158.8	85.1±13.4	107.7±37.1	73.06±52	2.1±1.4
P group						
Baseline	108.7±52.3	279.7±117.5	89.0±15.8	116.1±27.8	108.4±96.9	3.7±4.4
6 months			84.1±8.7			
12 months	95.8±44.4	266.5±114.4	94.2±12.6	117.8±37.1	76.6±43.8	2.5±1.4
P values						
Time	NS	NS	NS	NS	0.03	0.001
Interaction	0.009	NS	NS	NS	NS	NS

Results (mean±s.d.) of repeated measures ANOVA are reported. Significant differences ($P < 0.05$).

oral estrogens. It is well known that the effects on TG levels are dependent upon the route of estrogen administration (39).

Macrophage endothelial accumulation is associated with increased plasma concentrations of fibrinogen and C-reactive protein (40, 41), both markers of inflammation that are considered early signs of atherosclerosis (42, 43) and predictive factors for ischemic heart disease (37, 44). Despite these facts and the evidence of significantly elevated levels of C-reactive protein in women with polycystic ovarian syndrome (45), few studies have addressed the effects of androgen administration on these markers in women. Buckler *et al.* found no changes in fibrinogen, C-reactive protein and clotting factors in women treated with low doses of subcutaneous T implants for at least 2 years (46). Basaria *et al.*, however, described a significant increase in fibrinogen levels from baseline with the addition of MT (2.5 mg) to esterified estrogen, observed to a lesser extent with estrogen replacement in postmenopausal women (35). In the present work, we demonstrated a neutral effect of oral MT (1.25 mg) and percutaneous E₂ on C-reactive protein levels, and a significant decline in plasma fibrinogen levels with both therapies, suggesting a favorable estrogenic effect on this latter parameter that was preserved in women treated with low doses of MT.

The significant and similar reductions of systolic and diastolic blood pressure observed in our trial could also be attributed to parenteral estrogen replacement, an effect which was previously recorded in women with transdermal E₂ therapy (47). In concordance with earlier data (29), these results suggest that low-doses of oral MT have no adverse effects on blood pressure in postmenopausal women. An increased systolic blood pressure and rate pressure product at rest, however, was reported in women during oral esterified estrogens combined with MT replacement (48).

Relatively little is known regarding the influence of androgens on blood pressure. It has been suggested that testosterone elevates blood pressure by inducing catecholamine synthesis (49), or increasing circulating

levels of homocysteine (50), neuropeptide Y (51) and endothelin-1 (50) and the volume reabsorption rate of the proximal renal tubule (52). In contrast, direct vasodilatory and vasorelaxatory effects of testosterone are recognized in animals (53) and man, including postmenopausal women (31).

Perhaps the main findings in the present report are related to body composition. MT treatment produced a slight overall weight gain, a nonsignificant but relatively greater loss of subcutaneous abdominal fat mass and an accumulation of visceral fat, as determined by CT. Studies with androgen administration in postmenopausal women have shown improvements in body composition as assessed by photon absorptiometry, with indications of decreased fat body mass and increased lean body mass during therapy, the latter explaining the increases in body weight and BMI (6, 25, 54–56). However, only deep abdominal adipose tissue is clearly related to metabolic complications, and CT or magnetic resonance image scanning are the only available methods to distinguish visceral fat precisely (57).

Female central obesity has already been positively correlated with endogenous androgen levels (58–61). However, few studies have examined the effects of administering androgenic compounds to abdominal fat by CT. In a controlled study, Lovejoy *et al.* tested the effects of nandrolone decanoate in obese postmenopausal women during a 9-month clinical trial (61) with findings very similar to ours. They noted an increase in intra-abdominal fat and a loss of subcutaneous abdominal fat in the nandrolone-treated group, resulting in an increase in the ratio between these two fat depots in the abdomen.

The influence of androgen status on the regulation of lipolysis is not completely understood. Gender and regional differences, probably related to the number or activity of beta-adrenergic receptors and inhibitory alpha-adrenergic receptors, are suggested explanations (62). In male rats, testosterone stimulates catecholamine-induced lipolysis by increasing the

number of beta-adrenoceptors as well as the activity of adenylate cyclase (63). In a recent study, testosterone decreased catecholamine-induced lipolysis in subcutaneous abdominal fat cells while not affecting lipolysis in visceral fat cells of hyperandrogenic women (64).

One-year combined MT administration did not negatively influence glucose metabolism parameters tested in the present study. Fasting glucose and glucose tolerance were unchanged, while fasting insulin and insulin resistance were significantly reduced in the two groups, as might be expected during estrogen replacement (65–67). However, it is interesting to note that the improvements in insulin parameters appear to be greater in the estrogen-only group, suggesting some attenuating effect of MT on reducing insulin levels. Similar results were reported with a short-term MT administration (5 mg for 10–12 days) to regularly menstruating women studied by hyperglycemic and euglycemic hyperinsulinemic clamp techniques. The authors showed no significant androgen effects on fasting levels of glucose or insulin; nevertheless, the insulin response to this hyperglycemic challenge was slightly greater during MT treatment, providing direct evidence of a relationship between hyperandrogenemia and insulin resistance (68).

According to current research, this finding might be explained by the slight weight gain registered in the MT group, but it can also originate from visceral fat accumulation. The absolute volume of visceral adipose tissue appears to be a major determinant of insulin resistance in women (69), and it is closely associated with type 2 diabetes mellitus (70). Moreover, in a prospective study, greater visceral adiposity preceded the development of type 2 diabetes, an effect independent of family history, fasting insulin, insulin secretion, glycemia and total adiposity (71). Thus, further long-term studies in postmenopausal women are required in order to confirm the increases in the intra-abdominal fat mass during MT treatment, and to investigate the relationship between this increase and insulin resistance.

In conclusion, our study suggests that the combination of low-dose oral MT and percutaneous E₂, for 1 year, does not result in significant increase of cardiovascular risk factors. Although MT adversely affected HDL cholesterol and enhanced visceral fat, there were no detrimental effects on glucose metabolism, other lipids, inflammatory markers or blood pressure. This regimen can be recommended for symptomatic postmenopausal women with decreased libido, although it seems prudent to perform baseline and follow-up lipid profiles and assessment of body composition, especially in those at high risk of cardiovascular disease.

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