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Original Article

The Effects of Tibolone on Risk Factors of Cardiovascular Disease in Menopausal Women

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Abstract

Background: Tibolone is frequently used as a hormone-like alternative to traditional HRT. Therefore, it is valuable to compare the effects of tibolone and HRT on cardiovascular diseases risk factors.

Methods: A total of 156 healthy non-surgical postmenopausal women were included in an open randomized study. They were assigned to receive daily 2.5 mg tibolone plus one Cal+D (500 mg calcium and 200 IU vitamin D) tablet (n=52), 0.625 mg conjugated equine estrogen and 2.5 mg medroxy progesterone (CEE/MPA) plus one Cal+D tablet (n=52), or one Cal+D tablet (n=52). The latter group was used as control. The women were followed for six months. The body mass index (BMI), blood pressure (BP), serum levels of high density lipoprotein (HDL), low-density lipoprotein (LDL), and triglyceride (TG), C reactive protein (CRP), sex hormone binding globulin (SHBG), free testosterone index (FTI), and free estradiol index (FEI) were determined before and after the interventions.

Results: Compared to baseline values, BMI, BP, CRP, SHBG, HDL, FTI, and FEI were significantly different after the treatments in the tibolone and CEE/MPA receiving groups ($P<0.05$), but not in the control group. Moreover, serum LDL and TG levels changed significantly after the treatments in all the three groups ($P<0.01$). There were significant differences between the tibolone and CEE/MPA groups in terms of CRP, LDL, HDL, TG, SHBG, FTI, FEI and BP ($P<0.01$).

Conclusion: Tibolone may be considered an alternative to conventional HRT in postmenopausal women. However, clinical recommendations regarding the effects of tibolone on cardiovascular outcome needs further studies.
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Keywords ● C reactive protein ● hormone replacement therapy ● lipid ● sex hormone-binding globulin ● tibolone

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Introduction

The cessation of ovarian function at the time of menopause and consequent reduction in circulating estrogens result in a wide range of health problems, including vasomotor symptoms, genital atrophy, osteoporosis and detrimental cardiovascular changes.[1-3] Although hormone replacement therapy (HRT) offers a number of significant short-term benefits such as the relief of menopausal symptoms including hot flushes and vaginal

dryness and long-term ones such as the reduction in risk for osteoporosis and fractures, and possibly cardiovascular diseases and cognitive decline, its use is often problematic by poor compliance.[1-4] Women are often different in terms of tolerating the HRT side effects, particularly bleeding, and breast tenderness and pain, and they frequently fear the HRT-induced weight gain and breast cancer.[5-9] However, many women estimate their personal risk for major health problems erroneously, and are unaware of the long-term benefits of HRT. Thus, an ideal HRT would provide relief from vasomotor symptoms and vaginal dryness, improvement in libido and cognition, and protection against bone loss and cardiovascular disease, while avoiding endometrial and breast stimulation.

Tibolone is a steroid compound that is structurally related to 19-nortestosterone derivatives, such as norethisterone. Following oral administration, tibolone is metabolized to three active agents: 3-alpha hydroxyl-tibolone; 3-beta-hydroxy-tibolone; and Δ^4-tibolone. Preclinical data indicate that tibolone and its metabolites have estrogenic, progestogenic or androgenic activity, depending primarily on the target tissue involved.[1-4] For this reason, it is said that tibolone is a compound with tissue-specific activity.[1-4] Tiboline, is a frequently-used hormone-like alternative to traditional HRT, therefore, it is valuable to examine the influence of tibolone on the risk factors of cardiovascular diseases.

The objective of the present study was to compare the effects of tibolone and HRT on the risk factors of cardiovascular diseases.

Materials and Methods

The study, an open single blind randomized design, was approved by the Ethics Committee of the Tarbiat Modares University. The protocol of the study and the risks involved were explained to all participants, and informed consent was obtained.

Since there was no information relevant to this subject in the literature a pilot study was undertaken. Then assuming a β value of 0.1 and an alpha value of 0.05 to detect a 17 mg/dl difference in serum LDL levels between the tibolone (SD=26) and HRT (SD=23) groups, a sample size of 48 patients for each arm was calculated. To allow for possible losses to follow-up, a total of 156 healthy non-surgical postmenopausal women (age range 45-60), whose last menopausal period was more than one year earlier and their plasma levels of 17-ß-estradiol were <35 pg/ml were included. None of the subjects had taken any cholesterol-lowering agent, estrogen therapy, antioxidant vitamin supplements during the preceding 6 months. Patients with contraindications to HRT, dyslipidemia [cholesterol (CHOL) >250 mg/dl and/or triglyceride (TG) >150 mg/dl] were excluded. Also, none of the participants was a smoker, or had diabetes, previous angina or hypertension. Patients were randomly allocated into three groups of A, B or C, according to a computer-generated list of random numbers. Subjects of group A (n=52) were assigned to receive 2.5 mg tibolone (Tibofem, CIPLA LTD.) plus one Cal+D tablet (500 mg calcium and 200 IU vitamin D) daily. Women in groups B (n=52) were allocated to receive 0.625 mg conjugated equine estrogen and 2.5 mg medroxy progesterone (CEE/MPA) plus one Cal+D tablet daily. Subjects of group C did receive only one Cal+D tablet a day, and served as a control group. The participants were treated and followed up for six months. Of the initial 156 subjects, 6 withdrew from the study before the final analysis. The main reasons for dropping out were fear of breast cancer in one woman, breast tenderness in two subjects, and abnormal vaginal bleeding in three women. Finally, a total of 50 women in each group completed the study.

The serum levels of lipids including CHOL, high-density lipoprotein cholesterol (HDL), low density lipoprotein (LDL), TG, and also C-reactive protein (CRP), sex hormone binding globulin (SHBG), free testosterone index (FTI =testosterone × 100/SHBG), free estradiol index (FEI=estradiol × 100/SHBG) were determined before and after the interventions. In addition, a questionnaire was completed by all of the subjects before and after the treatments for probable drug side effects. Also, the blood pressure and body mass index (BMI) were recorded before and after the treatments. Blood samples were obtained between 10:00 and 12:00 h after an overnight fast. Laboratory measurements were carried out on the day of obtaining blood samples. The measurement of serum CRP levels was done using Immun Turb (Biosystems kit). Serum concentrations of CHOL and TG were determined by enzymatic methods (CHOL–PAP and TGO-PAP methods; Thechnicon Instr., NY). The HDL fraction was separated by the Mg^{2+}-phosphotungtic acid precipitation technique followed by enzymatic determination of CHOL. Plasma levels of LDL-CHOL were estimated according to Friedwald's procedure because the TG levels were below 300 mg/dl in all of the patients. The measurement of SHBG,

estradiol, and testosterone was done by ELISA (IBL, DRG and DRG Kits, respectively). Samples were handled in identical and blind fashion throughout the study. Then, they were analyzed in triplicate and in random orders so as to reduce bias and inter assay variations. All the measurements were repeated after 6 months of entering into the study. The physician who collected and analyzed the data did not know which subjects were placed in which group.

Statistical Analysis

The comparison of lipid levels among the three groups was prospectively designated as the primary end point. All other comparisons were considered secondary end points.

The data from the 3 groups, presented as mean±SD, were analyzed using one way Analysis of Variance (ANOVA). Where a significant difference was obtained with ANOVA, the source of difference was located using Tukey test for pairwise comparisons. Within group comparisons between the baseline and final data were performed using pair t test. The frequencies of gravidity were compared using Chi-square test. A p value of ≤0.05 was considered statistically significant. All confidence intervals were calculated at the 95 percent level.

Results

The baseline clinical and laboratory data including age, last menopausal age, gravidity, BMI, blood pressure, CRP, lipid profile, SHBG, FTI, and FEI were not significantly different among the three groups. After 6 months of treatment, there were significant differences in CRP, LDL, HDL, TG, SHBG, FTI, FEI levels and blood pressure of groups receiving tibolone and CEE\MPA. Moreover, there were significant (P<0.05) differences between the values of CRP, HDL, TG, SHBG and FTI from the HRT and tibolone groups and those of the control group. In addition, there was no significant difference between the serum levels of LDL from Cal+D and tibolone-treated groups (tables 1, 2).

There was no significant difference between baseline and final values of BMI, blood pressure, CRP, SHBG, HDL, FTI, and FEI in the control group. However, the values of BMI, blood pressure, CRP, SHBG, HDL, FTI, and FEI after the treatments in the tibolone and CEE\MPA groups were significantly (P≤0.05) different from those of the respective baselines. Moreover, serum levels of LDL and TG of all 3 groups after treatment were significantly (P<0.01) different from those of respective baseline values (tables 1, 2)

Discussion

Several prospective studies suggest that hormone therapy decreases the risk of coronary artery diseases in relatively young and healthy postmenopausal women.[5-7] The use of androgens in postmenopausal women are advised for vasomotor symptoms resistant to conventional HRT, postmenopausal women experiencing low libidio, osteoporosis in women treated adequately with other measures, loss of sense of well-being and cognitive functions.[8,9] On the other hand, when the adding of

Table 1: The values (mean±SD) of characteristics of participating women before and after treatment with Tibolone (group A), CEG/MPA (group B), or calcium-D only (group C).

	Group A (n:50)			Group B (n:50)			Group C (n:50)			P** value		
	Before	After	P*	Before	After	P*	Before	After	P*	(A,B)	(A,C)	(B,C)
Age (years)	51.78±3.29			51.58±2.82			52.52±4.06			0.95	0.52	0.35
Menopausal age (years)	49.54±2.81			48.84±2.36			49.06±3.16			0.95	0.74	0.89
Gravidity number (%)										0.28	0.43	0.95
< 3	9(18%)			6(12%)			7(14%)					
3-5	41(82%)			44(88%)			43(86%)					
BMI	27.74±3.59	28.01±3.69	0.019	28.50±3.80	29.16±3.70	<0.001	28.39±4.51	28.51±4.52	0.9	0.32	0.81	0.69
Systolic blood pressure (mmHg)	122±8.51	117.50±8.99	<0.001	122.10±9.48	126.60±8.83	<0.001	120.10±9.66	120.70±9.94	0.76	0.000	0.19	0.005
Diastolic blood pressure (mmHg)	79.30±6.62	77.78±6.90	0.003	79.25±8.88	82.30±6.64	<0.001	77.20±8.09	77.60±8.40	0.82	0.007	0.9	0.005

* indicate p values of paired t tests. ** indicate p values of ANOVA followed by Tukey test for after treatments values and for, χ^2 test used for the comparison of gravidity frequencies.

Table 2: The values (mean±SD) parameters measured for participating women before and after treatment with Tibolone (group A), CEG/MPA (group B), and calcium-D only (group C).

	Group A (n:50)			Group B (n:50)			Group C (n:50)			P** value		
	Before	After	P*	Before	After	P*	Before	After	P*	(A,B)	(A,C)	(B,C)
CRP(mg/l)	2.23±1.04	2.87±1.12	<0.001	1.80±1.08	4.03±1.26	<0.001	2.16±1.24	2.05±1.01	0.63	<0.001	0.001	<0.001
HDL(mg/dl)	44.40±9.42	33.64±9.28	<0.001	44.98±12.03	63.34±11.18	<0.001	46.02±10.53	44.62±10.77	0.51	<0.001	<0.001	<0.001
LDL(mg/dl)	121.92±18.08	116.50±18.20	<0.001	115.23±18.12	89.64±17.13	<0.001	122.38±15.29	118.07±16.09	<0.01	<0.001	0.65	<0.001
TG(mg/dl)	123.41±41.84	75.30±31.52	<0.001	122.30±35.36	181.58±37.57	<0.001	132.75±38.19	126.12±39.70	<0.001	<0.001	<0.001	<0.001
SHBG(nmol/l)	41.69±25.46	22.45±12.42	<0.001	43.89±18.36	75.86±37.29	<0.001	44.25±23.82	40.18±18.90	0.08	<0.001	0.001	<0.001
Free Estradiol Index	0.31±0.36	0.59±0.59	0.001	0.30±0.35	0.28±0.31	0.49	0.33±0.38	0.33±0.38	0.34	<0.001	0.011	0.47
FreeT-estestrone Index	0.05±0.07	0.70±0.30	<0.001	0.03±0.02	0.04±0.03	<0.01	0.05±0.08	0.04±0.01	0.23	<0.001	<0.001	0.95

* indicates p values for paired t test. ** indicate p values for ANOVA followed by Tukey test on after treatment values.

androgens is considered, cardiovascular issues and lipids are major concerns. Tibolone, a synthetic steroid with estrogenic, androgenic and progestogenic properties, relieves climacteric symptoms, prevents postmenopausal bone loss, and helps depression and libidio.[10] Furthermore, compared with HRT, tibolone has no effect on breast cancer incidence, and does not promote endometrial hyperplasia or rare vaginal bleeding.[3,4] Animal studies have demonstrated that the use of tibolone results in significantly less atherosclerosis than does the use of placebo, or at least, that the use of tibolone was associated with no increase in coronary atherosclerosis relative to control despite a marked reduction of HDL cholesterol levels.[11,12]

The recognition that atherosclerosis is an inflammatory process has led to the evaluation of several plasma markers of inflammation as potential tools for the prediction of risks of coronary events. The measurement of markers of inflammation, in addition to standard screening of lipid levels, might provide a clinically useful method for improving the overall prediction of the risk of cardiovascular events.[13-18] In the present study, we conducted a prospective randomized control design involving 156 apparently healthy postmenopausal women over a follow-up period of six months to assess the risk of cardiovascular events associated with the baseline levels of lipids and CRP. We compared the mentioned parameters as well as BMI, blood pressure, SHBG, free testosterone index, and free estradiol index among women who received conventional HRT plus Cal+D, tibolone plus Cal+D, or Cal+D only as a control group. After six months of treatment, Cal+D had beneficial effects on the markers of cardiovascular diseases such as the reduction of LDL and TG. In a recent study, lipid profile was tested after a long-term calcium supplementation in women with low calcium intake. The study showed a significant decrease in LDL cholesterol levels, as well as an improvement in the total LDL and ratio of LDL to HDL cholesterol.[19] The present study used women who received only Cal+D as a control group, and found results contrary to our expectation. However, since Cal+D were administered to all groups, we can compare the effects of treatments on the cardiovascular risk factors among the three groups. As shown in the results, tibolone reduced LDL, HDL and TG, and increased CRP after six months. Also, HRT reduced LDL, and increased HDL, TG and CRP after the treatment. On the other hand, a comparison between tibolone and CEE\MPA groups showed that tibolone increased CRP level and decreased LDL level less than what CEE\MPA did. Also, in contrast to CEE\MPA, tibolone decreased TG. The comparison between tibolone and control groups showed that tibolone significantly reduced TG, HDL cholesterol and also TG/HDL ratio, which, in contrast to increased CRP, is a powerful predictor of insulin resistance and coronary heart disease. Although tibolone reduced LDL and TG, it increased CRP and decreased HDL. A comparison among the three groups showed that tibolone significantly reduced TG, HDL cholesterol and TG/HDL ratio, which is a powerful predictor of insulin resistance and coronary heart disease. Moreover, tibolone increased CRP levels less than that of HRT. In the present study, HRT, and tibolone increased the CRP levels, but the increase by tibolone was less remarkable. The results of the present study confirm the findings of a number of previous studies which have shown that tibolone increases CRP levels and decreases TG and HDL levels.[1-4,16-18] However, the effects of tibolone on other risk factors of atherosclerosis such as increasing fibrinolytic activities and improving flow-mediated brachial artery vasodilation is clearly beneficial, and might theoretically counterbalance the potentially negative effect of tibolone on HDL and CRP levels.[1,2] Epidemiological studies have consistently shown that elevated CRP is a risk factor for coronary heart disease, even in apparently healthy women.[20,21]

A recent study showed that CRP level increased after three years of HRT treatment in the WHI (women's health initiative) trial.[22] The finding that CRP has several important atherogenic properties and is an inflammation marker, indicates that an HRT and tibolone-induced CRP increases may result in the progression of atherogenesis.

Since it might be possible that the effects of tibolone on HDL cholesterol and the effects of HRT on TG are related to their effects on the steroid levels, the serum levels of SHBG, FTI and FEI were measured. The Majority of sex steroids, estradiol and testosterone are bound to SHBG, a protein carrier produced in the liver. High androgen level decreases SHBG, while high estrogen level increases it. Two previous studies have shown that systemic conventional HRT preparations increase SHBG and estradiol plasma levels, and decrease plasma androgen levels.[23-25] Also, tibolone was shown to decrease the level of SHBG, and substantially increases free testosterone.[25] Similarly, in the present study tibolone treatment

decreased the serum levels of SHBG, which might have led to the increase of serum levels of FTI and FEI. In contrast to tibolone, HRT therapy increased SHBG and decreased FTI. The effects of tibolone on HDL cholesterol and HRT on TG may be linked to their effects on SHBG, FTI and FEI.

Although, when compared to the baseline values, BMI in CEE/MPA and tibolone groups increased significantly after the treatments, they did not change significantly when they were compared with that of Cal+D treated group. Such a finding is similar to our previous finding about the changes of body composition in postmenopausal women who were taking tibolone.[26]

Conclusion

The findings of the present study indicate that systolic and diastolic blood pressures; the two important cardiovascular risk factors, decreased in the tibolone group, and increased in the HRT group. Such findings indicate that tibolone may be considered an alternative to traditional HRT in postmenopausal women. However, clinical recommendations regarding the effects of tibolone on cardiovascular outcome must await the performance of additional studies with clinical end points.

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Conflict of Interest: None declared

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