Bio-Medical Materials and Engineering 22 (2012) 179–185
DOI 10.3233/BME-2012-0705
IOS Press

Imbalance of testosterone/estradiol promotes male CHD development

Hong-Yun Zheng, Yan Li *, Wen Dai, Chuan-Dong Wei, Kai-Sheng Sun and Yong-Qing Tong
Department of Clinical Laboratory, Renmin Hospital of Wuhan University, Wuhan, China

Abstract. *Objective*: Testosterone is either neutral or has a harmful effect on the male cardiovascular system. But the role of imbalance of testosterone (T) and estrogen (E2) (T/E2 ratio) in male CHD has been less studied. This study was carried out with the purpose of evaluating the relationship between T/E2 ratio and CHD.

Methods: Fifty-five male CHD patients (aged 61.25 $\pm$ 3.44) and 60 age-matched controls (aged 59.54 $\pm$ 1.44) were selected in this research.

Results: Compared with control group, levels of both serum T and E2 decreased, but only E2 had statistical significance ($P = 0.001$). The normal testosterone (T)/estradiol (E2) ratio is 1.7 $\pm$ 0.12, but the ratio of T/E2 (3.28 $\pm$ 0.58) changed significantly in men with CHD group ($P < 0.05$). With the imbalance of T/E2 ratio in CHD group, we further used a linear and multiple regression methods to analyze the correlation between sex hormones and CHD risk factors. The results showed serum T was positively associated with TG ($r = 0.439$, $P < 0.01$) and D-dimer ($r = 0.258$, $P < 0.05$), but negatively associated with HDL-C ($r = -0.267$, $P < 0.05$) and Hs-CRP ($r = -0.214$, $P < 0.05$). However, E2 was highly positive associated with TG ($r = 0.783$, $P < 0.01$) and HDL-C ($r = 0.515$, $P < 0.01$), but was negative related with LDL-C ($r = -0.219$, $P < 0.05$), TC/LDL ($r = -0.236$, $P < 0.05$) and D-dimer. Multiple linear regression method also showed the same results between E2 and HDL-C ($P = 0.020$), LDL-C ($P = 0.000$), which showed E2's protective role in cases. However, T/E2's effect is more significative than E2's, and the values between T/E2 and index are HDL-C ($r = -0.624$, $P < 0.01$), LDL-C ($r = 0.348$, $P < 0.01$), TC/HDL ($r = 0.237$, $P < 0.05$), Hs-CRP ($r = 0.248$, $P < 0.05$) and D-dimer ($r = 0.249$, $P < 0.05$). Multiple linear regression method also showed the positive relationship between T/E2 and HDL-C ($P = 0.000$), D-dimer ($P = 0.000$), and negative relationships between T/E2 and TC ($P = 0.000$), TG ($P = 0.000$) or HDL/LDL ($P = 0.000$).

Conclusion: The balance of T/E2 ratio, rather than the absolute levels of androgens, is crucial in modulating the effect of androgens on CHD in males.

Keywords: Coronary heart disease, testosterone, estradiol, imbalance, men

1. Introduction

Compared with women of similar age, cardiovascular diseases (CVD) are more prevalent in men [1]. What's more, men present a greater incidence of coronary heart disease (CHD) [2]. Although androgens and estrogens are the main sex hormones found in men and women, respectively, it is unlikely that estrogens are important only in women, nor is it likely that it exert the same effects in men and women. Sex hormones exert metabolic [3] and hemodynamic effects [4] that might explain gender differences in the relevance of risk factors in determining cardiovascular risk. However, many studies are uncertain about the relationship between imbalance of sex hormones and occurrence of CHD.

Both estrogen and testosterone have important roles to play in individuals of either sex [5]. Testosterone metabolized by 5α-reductase to the more potent AR ligand, dihydrotestosterone (DHT) [6]. Al-

* Address for correspondence: Dr. Yan Li, Department of Clinical Laboratory, Renmin Hospital of Wuhan University, Wuhan, Hubei 430060, China. Tel.: +86 27 88041911-88258; E-mail: yanlitf@yahoo.com.cn.

ternatively, testosterone is metabolized by aromatase to the primary form of estrogen, 17β-estradiol [7]. Thus, in both males and females, the balance between testosterone and estrogen production through various life stages influences the function of cardiovascular system. However, the relationship between sex hormones and the cardiovascular system, in both men and women, remain controversial. *In vitro* studies, 17β estradiol at high local concentrations inhibits LDL oxidation and reduces cholesterol ester formation [8,9], but testosterone does not have the same effect [9], which could further increase the incidence of CHD. There was also a trend for men to have higher LDL-C concentrations than women and for women to have higher HDL-C concentrations than men. Experimental evidence suggests that androgen deficiency contributes to the onset and progression of CHD in men [10]. Androgen deficiency is associated with endothelial dysfunction [11], high glucose and adverse lipid profiles [12], inflammatory responses [13], altered smooth muscle and hypertension [14], which are associated with CHD. However, *in vivo*, androgens and estrogens have opposing actions on function of blood vessels [15]. For example, in ovariectomized female animals, estrogen replacement generally restores cerebrovascular function to that found in cerebral vessels from intact, untreated females [16–18]. However, in males, the situation is more complex. Testosterone replacement in castrated male rats does not restore cerebrovascular function [18,19]. Moreover, certain responses in arteries in males, such as nitric oxide dependent dilation, are reproduced by estrogen treatment of castrated males [18]. These findings suggest that active androgen metabolites, which may differ depending on the balance of androgen and estrogen in the cardiovascular system in males. So we think there must be a normal serum testosterone/estradiol (T/E2) balance in normal males. Once the T/E2 balance is broken, which may be contribute to the incidence of CHD.

In this work, we first want to observe the normal physiological T/E2 ratio in health old men, and then to explore the effect of imbalance of T/E2 on the CHD-related risk factors, such as serum lipids, inflammatory factors and D-dimer in men with CHD.

2. Subjects and methods

2.1. Subjects

Subjects were recruited from the Renmin Hospital of Wuhan University, Hubei Province in China from November 2010 to March 2011. The healthy old men in control group were selected from health center (age 45–60, $n = 60$). All of them had been excluded heart disease, diabetes mellitus, breast disease or other serious diseases. The similar aged men (age 45–60, $n = 55$) in case group were selected from department of cardiology.

2.2. Hormone assays

Samples were analyzed at the department of clinical laboratory of Renmin Hospital of Wuhan University on the ACS-180 automated analyzer (Bayer Diagnostics, France). Testosterone (T) and estradiol (E2) were measured by the method of competitive immunoassay and used the technology of direct chemiluminescent immunoanalysis. T assay range was 2–478 ng/dl. E2 was assayed using a rabbit anti-E2-6 ACS-180 immunoassay (the lower limit of detection to 1.0 pg/ml). T/(10 $\times$ E2) was used to make the resulting ratio [20].

2.3. Biochemical assays

Serum Total cholesterol (TC), HDL-C, LDL-C, triglycerides (TG), glucose, C-reactive protein (CRP) were assessed by an automatic biochemistry analyzer AU5400 (Olympus, Japan). TC, TG

and HDL-C were analyzed by enzymatic methods, and LDL-C was calculated using the equation (LDL-C = (cholesterol-triglycerides)/5-HDL-cholesterol) [21]. Glucose was determined by hexokinase (HK) method. CRP was quantified using an immune turbidmetric analysis method. D-dimer was measured by automated coagulation instrument Sysmex CA-7000 (Sysmex, Japan) using an immune turbidmetric analysis method. Linear range: TC: 0.5–18 mM; TG: 0.1–11.3 mM; HDL-C: 0.05–4.65 mM; LDL-C: 0.26–10.3 mM; glucose: 0.0–22.24 mM; CRP: the lower limit was 0.03 mg/dl; D-dimer: 0–0.3 μg/ml.

2.4. Statistical analysis

Statistical analysis was performed by use of SPSS 13.0. All data were expressed as mean ± SEM. Multiple linear regression and Pearson correlations were used. Statistical significance was set at the 0.05 level ($^{*}P < 0.05$, $^{**}P < 0.01$).

3. Results

3.1. Levels of serum testosterone, estrogen and CHD-related values

Mean age of control group is 59.54 ± 1.44, with a mean body mass index (BMI) 24.28 ± 0.76, and there were no differences between old male patients with CHD and control males in terms of mean age ($P = 0.332$) and BMI ($P = 0.344$). However, sex hormone as well as its CHD-related values had huge differences between controls and cases. As shown in Table 1, in control group, the average levels of testosterone (T) and estradiol (E2) are 611.42 ± 49.24 ng/dl and 36.35 ± 1.85 pg/ml respectively, however, the levels reduced to 522.44 ± 48.74 ng/dl and 20.73 ± 3.36 pg/ml in men with CHD group. Compared with control group, E2 levels decreased significantly ($P = 0.001$), but there was no statistically significance between the T levels' ($P = 0.214$). The normal T/E2 ratio is 1.70 ± 0.12, but its changed significantly (3.28 ± 0.58) in men with CHD group ($P = 0.020$). In the aspect of lipoprotein

Table 1
The average serum levels of sex hormones and CHD-related risk factors

Character	Controls (total $n = 60$)	Cases (total $n = 55$)	P-value
Age	59.54 ± 1.44	61.25 ± 0.99	0.332
BMI	24.28 ± 0.76	25.18 ± 0.58	0.344
T	611.42 ± 49.24	522.44 ± 48.74	0.214
E2	36.35 ± 1.85	20.73 ± 3.36	0.001
T/E2	1.70 ± 0.12	3.28 ± 0.58	0.020
TC	4.24 ± 0.17	4.31 ± 0.17	0.763
TG	0.85 ± 0.06	1.16 ± 0.12	0.035
HDL	1.33 ± 0.05	1.12 ± 0.05	0.009
LDL	2.40 ± 0.13	2.60 ± 0.15	0.319
TC/HDL	3.22 ± 0.15	1.06 ± 0.12	0.000
HDL/LDL	0.53 ± 0.03	0.50 ± 0.05	0.675
HsCRP	2.12 ± 0.18	9.03 ± 2.37	0.011
D-dimer	0.072 ± 0.014	0.21 ± 0.15	0.000

Notes: All the values had been adjusted for age and BMI. P-values from t-test for independent samples.

profile, in cases group, the level of HDL-C decreased significantly ($P = 0.009$); TG increased significantly ($P = 0.035$), and LDL-C changed slightly ($P = 0.319$). But the ratio of TC and HDL (TC/HDL) was lower ($P = 0.000$) than the control's. Both of Hs-CRP and D-dimer levels increased sharply in male patients with CHD compared with healthy men ($P = 0.011$, $P = 0.000$).

3.2. Correlation of the serum sex hormones levels with CHD risk factors in cases

In order to explore the relationship between sex hormone and CHD-related risk factors, we conducted a linear analysis of the data in case group. As shown in Table 2, the results showed that average serum level of T was positive associated with TG ($r = 0.439$, $P < 0.01$) and D-dimer ($r = 0.258$, $P < 0.05$). On the other hand, T was mainly negative associated with HDL-C ($r = -0.267$, $P < 0.05$) and Hs-CRP ($r = -0.214$, $P < 0.05$). Serum E2 was highly positive associated with TC ($r = 0.263$, $P < 0.05$), TG ($r = 0.783$, $P < 0.01$) and HDL-C ($r = 0.515$, $P < 0.01$), but was negative related with LDL-C ($r = -0.219$, $P < 0.05$) and TC/LDL ($r = -0.236$, $P < 0.05$). The ratio of T/E2 was inversely related with TC ($r = -0.376$, $P < 0.01$), and HDL-C ($r = -0.624$, $P < 0.01$), but positively related with LDL-C ($r = 0.348$, $P < 0.01$), TC/HDL ($r = 0.237$, $P < 0.05$), Hs-CRP ($r = 0.248$, $P < 0.05$) and D-dimer ($r = 0.249$, $P < 0.05$).

3.3. The relationship of imbalance of T/E2 with CHD high risk factors-lipoprotein profile in cases

As shown in Table 3, T was significantly positively associated with TG ($P = 0.030$) and LDL-C ($P = 0.009$) in cases group after adjusting for age and BMI. And there were no significant relationship

Table 2

Correlation between sex hormones and lipoprotein in patients with CHD

	T	E2	T/E2
TC	−0.174	0.263*	−0.376**
TG	0.439**	0.783**	−0.110
HDL	−0.267*	0.515**	−0.624**
LDL	−0.178	−0.219*	0.348**
TC/HDL	0.088	−0.236*	0.237*
HDL/LDL	0.048	0.142	−0.038
HsCRP	−0.214*	0.059	0.248*
D-Dimer	0.258*	−0.008	0.249*

Notes: Linear analysis of the data in case group, all values had been adjusted for age and BMI. $^{*}P < 0.05$; $^{**}P < 0.01$.

Table 3

Relationship of serum sex hormones levels to lipoprotein profile in patients with CHD

Sex hormone	TC		TG		HDL		LDL	
	β	P-value	β	P-value	β	P-value	β	P-value
T	0.426	0.197	2.736	0.030	−1.413	0.074	1.264	0.009
E2	0.564	0.007	−0.458	0.549	1.147	0.020	−1.536	0.000
T/E2	−1.418	0.000	−4.720	0.000	3.005	0.000	0.399	0.033

Note: Multiple linear regression analysis, generalized estimating equation models adjusted for age and BMI.

between T and TC or HDL. In contrast, E2 mediated lipids profile more effectively in old men with CHD. E2 had a significantly positive association with TC ($P = 0.007$) and HDL-C ($P = 0.020$) and a negatively association with LDL-C ($P = 0.000$). According to the result, T/E2 had a strongest association with lipids. The positive relationship was observed between T/E2 and HDL ($P = 0.000$) or LDL ($P = 0.033$) and negative relationships between T/E2 and TC or TG, with both P-values were 0.000.

3.4. The relationship of imbalance of T/E2 with other CHD risk factors in cases

As shown in Table 4, we could see T was significantly negatively associated with TC/HDL ($P = 0.010$), but it was not associated with HDL/LDL ($P = 0.102$) in case group after adjusting for age and BMI. However, the effect of E2 is completely different. E2 had a moderately positive association with TC/HDL and had a negatively association with HDL/LDL ($P = 0.000$). In addition, we also observed some other interesting CHD-related risk factors, such as inflammatory factor-Hs-CRP and thrombosis associated risk factor-D-dimer. From the Table 4, we can see T was significantly negatively associated with D-dimer ($P = 0.002$), and was slightly positively associated with Hs-CRP. Nevertheless, E2 was significantly negatively associated with both Hs-CRP ($P = 0.000$) and D-dimer ($P = 0.000$) in case group. According to the results, T/E2 had a strongest positively association with TC/HDL ($P = 0.000$) and D-dimer ($P = 0.000$). There is no strong relation between T/E2 and Hs-CRP ($P = 0.120$).

4. Discussion

A common explanation for the greater incidence of CHD in men when compared to women could be the high levels of serum testosterone in men, which suggests testosterone could promote atherosclerosis or estrogen could protect men from CHD [22]. Whether androgens affect cardiovascular disease adversely remains a controversial issue, with some data pointing to a deleterious effect of androgens on lipid profiles [23], but some other studies revealed androgens exerted possible benefits on cardiovascular function. Many studies examined the relationship between endogenous testosterone and CHD incidence in men, but failed to get a clear association [24]. Cauley et al. [25] found similar testosterone concentrations in 163 men who had coronary events compared with matched controls. High-density lipoprotein cholesterol (HDL-C) is an anti-atherogenic factor, but low-density lipoprotein cholesterol (LDL-C) is a pro-atherogenic risk factor, which is quite harmful to vascular system. D-dimer could promote thrombosis. Most of the researches reported E2 have beneficial effects on the male cardiovascular function by inhibiting proliferation of vascular smooth muscle cells [26] and increasing HDL-C [27] and nitric oxide synthase activity (NOS), thus promoting vasodilatation [28]. Estrogen should have an important role to play in individuals of old men, however, with different levels of endogenous androgens, the effect

Table 4
Relationship of serum sex hormones levels to some other cardiovascular risk factors in patients with CHD

Sex hormone	TC/HDL		HDL/LDL		HsCRP		D-dimer	
	β	P-value	β	P-value	β	P-value	β	P-value
T	−3.725	0.010	1.121	0.102	0.073	0.326	−0.269	0.002
E2	0.392	0.651	−2.093	0.000	−6.161	0.000	−19.788	0.000
T/E2	5.237	0.000	−1.940	0.000	0.045	0.120	1.030	0.000

Note: Multiple linear regression analysis, generalized estimating equation models adjusted for age and BMI.

of estrogen on cardiovascular system also different. So we suppose the balance of T/E2 ratio will be exerting more effect on CHD-related risk factors.

An important first step is to make sure whether the cardiovascular function in males is subject to the imbalance of testosterone and estrogen effects. In this study (Table 1), with no differences of ages and BMI in control and cases, compared with control, we found the average level of E2 decreased significantly ($P = 0.001$), but there was no statistically significance between the T levels' ($P = 0.214$), which indicated the patients with severe CAD had lower E2, but only slightly lower testosterone when compared with the control. The normal T/E2 ratio was 1.70 ± 0.12, but thus was broken in men with CHD group with changed ratio (T/E2: 3.28 ± 0.58). Moreover, in cases group, the level of HDL-C ($P = 0.009$) and TC/HDL (0.000) decreased significantly; TG, Hs-CRP and D-dimer levels increased sharply in male patients with CHD compared with healthy men increased significantly ($P = 0.035$), but LDL-C changed slightly ($P = 0.319$). Inflammation, such as Hs-CRP has been used as an important independent predictor of CHD [29].

Some study found a neutral effect [24,30,31]of T and E2 levels with CHD. In the Tromso study [31], men aged 59.6 ± 10.2 years followed for up to 13 years, total and free T and E2 levels were not associated with occurrence of CHD. In order to see the correlation between sex hormones and CHD-associated risk factors, we conducted a linear analysis of the data in case group. Table 2 results showed that T level was positively associated with TG ($r = 0.439$, $P < 0.01$) and D-dimer ($r = 0.258$, $P < 0.05$), but negatively associated with HDL-C ($r = -0.267$, $P < 0.05$) and Hs-CRP ($r = -0.214$, $P < 0.05$). HDL-C is an anti-artherosclerosis factor, but LDL-C is quite harmful to vascular system. D-dimer could promote thrombosis. T was positive associated with TG and D-dimer, but negative associated with HDL-C, which means its harmful effect on vascular function. However, E2 and indexes changed absolutely. E2 was highly positive associated with TG ($r = 0.783$, $P < 0.01$) and HDL-C ($r = 0.515$, $P < 0.01$), but was negative related with LDL-C ($r = -0.219$, $P < 0.05$), TC/LDL ($r = -0.236$, $P < 0.05$) and D-dimer. From this result, we could see E2 played a more important role in protection old men from CHD. The relationship between T/E2 and risk factors is similar to E2's, but it is more significative than E2's effect on HDL-C ($r = -0.624$, $P < 0.01$), LDL-C ($r = 0.348$, $P < 0.01$), Hs-CRP ($r = 0.248$, $P < 0.05$) and D-dimer ($r = 0.249$, $P < 0.05$). Next, we used multiple linear regression to further analyse the results. From the Tables 3 and 4, we got the same results about the relationship between T, E2 or T/E2 with CHD risk factors. This result is really very exciting, which suggested neither single T nor E2 could not take complete responsibility for CHD. Our results showed that imbalance of serum T/E2 could be the most important reason for CHD in old men.

In conclusion, the present study found serum T/E2 balance is more useful index to reflect the relationship between sex hormones and CHD incidence. Imbalance of testosterone and estrogen could lead to hypercholesterolemia, hypertriglyceridemia, hyperglycemia, and hyperuricemia, which contributed to CHD. The T/E ratio, rather than the absolute levels of androgens or estrogen, is crucial in modulating the effect of androgens on CHD in males, which provided new ideas for diagnose and treatment of CHD patients in males.

References

[1] D.J. Lerner and W.B. Kannel, Patterns of coronary heart disease morbidity and mortality in the sexes: a 26-year follow-up of the Framingham population, *Am. Heart J.* **111**(2) (1986), 383–390.

[2] E. Wehr, S. Pilz, B.O. Boehm et al., Low free testosterone is associated with heart failure mortality in older men referred for coronary angiography, *Eur. J. Heart Fail.* **13**(5) (2011), 482–488.

[3] C. Vitale, M.E. Mendelsohn and G.M. Rosano, Gender differences in the cardiovascular effect of sex hormones, *Nat. Rev. Cardiol.* **6**(8) (2009), 532–542.
[4] J. Sagrista-Sauleda, J. Angel, A. Sambola et al., Hemodynamic effects of volume expansion in patients with cardiac tamponade, *Circulation* **117**(12) (2008), 1545–1549.
[5] E.R. Simpson, Sources of estrogen and their importance, *J. Steroid Biochem. Mol. Biol.* **86**(3–5) (2003), 225–230.
[6] J. Fujimoto, M. Nishigaki, M. Hori et al., The effect of estrogen and androgen on androgen receptors and mRNA levels in uterine leiomyoma, myometrium and endometrium of human subjects, *J. Steroid Biochem. Mol. Biol.* **50**(3,4) (1994), 137–143.
[7] S.E. Bulun, S. Sebastian, K. Takayama et al., The human CYP19 (aromatase P450) gene: update on physiologic roles and genomic organization of promoters, *J. Steroid Biochem. Mol. Biol.* **86**(3–5) (2003), 219–224.
[8] A. Negre-Salvayre, M.T. Pieraggi, L. Mabile et al., Protective effect of 17 beta-estradiol against the cytotoxicity of minimally oxidized LDL to cultured bovine aortic endothelial cells, *Atherosclerosis* **99**(2) (1993), 207–217.
[9] L.A. Huber, E. Scheffler, T. Poll et al., 17 beta-estradiol inhibits LDL oxidation and cholesteryl ester formation in cultured macrophages, *Free Radic. Res. Commun.* **8**(3) (1990), 167–173.
[10] Third Report of the National Cholesterol Education Program (NCEP), Expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (adult treatment panel III) final report, *Circulation* **106**(25) (2002), 3143–3421.
[11] D. Javeshghani, M.R. Sairam, E.L. Schiffrin et al., Increased blood pressure, vascular inflammation, and endothelial dysfunction in androgen-deficient follitropin receptor knockout male mice, *J. Am. Soc. Hypertens.* **1**(5) (2007), 353–361.
[12] A.M. Traish, R. Abdou and K.E. Kypreos, Androgen deficiency and atherosclerosis: The lipid link, *Vascul. Pharmacol.* **51**(5,6) (2009), 303–313.
[13] A.M. Traish, R.J. Feeley and A. Guay, Mechanisms of obesity and related pathologies: androgen deficiency and endothelial dysfunction may be the link between obesity and erectile dysfunction, *FEBS J.* **276**(20) (2009), 5755–5767.
[14] A.M. Traish, F. Saad, R.J. Feeley et al., The dark side of testosterone deficiency: III. Cardiovascular disease, *J. Androl.* **30**(5) (2009), 477–494.
[15] D.N. Krause, S.P. Duckles and D.A. Pelligrino, Influence of sex steroid hormones on cerebrovascular function, *J. Appl. Physiol.* **101**(4) (2006), 1252–1261.
[16] N. Geary, The effect of estrogen on appetite, *Medscape Womens Health* **3**(6) (1998), 3.
[17] A.M. McNeill, N. Kim, S.P. Duckles et al., Chronic estrogen treatment increases levels of endothelial nitric oxide synthase protein in rat cerebral microvessels, *Stroke* **30**(10) (1999), 2186–2190.
[18] G.G. Geary, D.N. Krause and S.P. Duckles, Estrogen reduces mouse cerebral artery tone through endothelial NOS- and cyclooxygenase-dependent mechanisms, *Am. J. Physiol. Heart Circ. Physiol.* **279**(2) (2000), H511–H519.
[19] A. Razmara, D.N. Krause and S.P. Duckles, Testosterone augments endotoxin-mediated cerebrovascular inflammation in male rats, *Am. J. Physiol. Heart Circ. Physiol.* **289**(5) (2005), H1843–H1850.
[20] M.R. Sowers, J. Randolph, Jr., M. Jannausch et al., Levels of sex steroid and cardiovascular disease measures in premenopausal and hormone-treated women at midlife: implications for the "timing hypothesis", *Arch. Intern. Med.* **168**(19) (2008), 2146–2153.
[21] W.T. Friedewald, R.I. Levy and D.S. Fredrickson, Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge, *Clin. Chem.* **18**(6) (1972), 499–502.
[22] E.Q. Callou de Sa, F.C. de Sa, R.D. Silva et al., Endogenous oestradiol but not testosterone is related to coronary artery disease in men sex hormones and coronary artery disease, *Clin. Endocrinol. (Oxf.)* **75**(2) (2011), 177–183.
[23] M.D. Schwarcz and W.H. Frishman, Testosterone and coronary artery disease, *Cardiol. Rev.* **18**(5) (2010), 251–257.
[24] J. Arnlov, M.J. Pencina, S. Amin et al., Endogenous sex hormones and cardiovascular disease incidence in men, *Ann. Intern. Med.* **145**(3) (2006), 176–184.
[25] J.A. Cauley, J.P. Gutai, L.H. Kuller et al., Usefulness of sex steroid hormone levels in predicting coronary artery disease in men, *Am. J. Cardiol.* **60**(10) (1987), 771–777.
[26] K. Sudhir and P.A. Komesaroff, Clinical review 110: Cardiovascular actions of estrogens in men, *J. Clin. Endocrinol. Metab.* **84**(10) (1999), 3411–3415.
[27] F.M. Sacks and B.W. Walsh, Sex hormones and lipoprotein metabolism, *Curr. Opin. Lipidol.* **5**(3) (1994), 236–240.
[28] P.W. Shaul, Novel role of estrogen receptors in vascular endothelium, *Semin. Perinatol.* **24**(1) (2000), 70–74.
[29] P.M. Ridker, M. Cushman, M.J. Stampfer et al., Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men, *N. Engl. J. Med.* **336**(14) (1997), 973–979.
[30] S. Turhan, C. Tulunay, S. Gulec et al., The association between androgen levels and premature coronary artery disease in men, *Coron. Artery Dis.* **18**(3) (2007), 159–162.
[31] T. Vikan, H. Schirmer, I. Njolstad et al., Endogenous sex hormones and the prospective association with cardiovascular disease and mortality in men: the Tromso study, *Eur. J. Endocrinol.* **161**(3) (2009), 435–442.
[32] V. Guarner-Lans, M.E. Rubio-Ruiz, I. Perez-Torres et al., Relation of aging and sex hormones to metabolic syndrome and cardiovascular disease, *Exp. Gerontol.* **46**(7) (2011), 517–523.