ATHEROESCLEROSIS RISK FACTORS IN WOMEN DURING MENOPAUSE
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Abstract
The loss of female sex hormones after menopause contributes to the striking increase in the incidence of cardiovascular morbidity and mortality. The present study was designed to evaluate some of the atherosclerotic risk factors: LDL-C, HDL-C and factor VII of coagulation, in women during menopause.

The study comprised a number of 77 women divided into three groups. The control group included 25 healthy women in their reproductive period. The perimenopausal group consisted of 27 women, with FSH level under 25mU/ml, and with anamnestic data of irregularity of menstrual cycle. The postmenopausal group encompassed 25 women regarding lack of cycle for more than 12 months. Hormone level was determined with RIA method. Lipid level was determined with standard colorimetric-spectrophotometric method and the concentration of f. VII was determined by method of plasma deficiency.

Statistical analysis has shown that there was a significant increase of LDL-C and factor VII in both perimenopausal and postmenopausal examinees in comparison with the control group (p<0.001).

This study favours the view that decrease in estradiol level and associated increase in LDL-CH and factor VII seen in perimenopausal and postmenopausal women may be responsible for the increased risk of atherosclerotic thromboembolic complications in women during the most vulnerable period of their life.

Key words: atherosclerosis, menopause.
hand, through ERα and ERβ, estrogens are involved in the proliferation of adipocytes, whereas their deprivation increases central obesity which is associated with a more atherogenic profile [9].

Estrogens have indirect protective effect on the blood vessels by regulating the ratio between serum lipids, especially HDL-CH/LDL-CH index that is an important predictor of coronary diseases. With menopause, the levels of total cholesterol and LDL-CH increase and the levels of HDL-CH decrease (especially HDL2 subfraction) [10]. There is clear evidence that high level of LDL-CH increases the risk of cardiovascular diseases, however HDL-CH is in reverse relation with the risk of cardiovascular diseases. Estrogens contribute to this concept probably through 2 basic mechanisms:

- The increase of HDL-CH synthesis;
- The decrease of HDL-CH catabolism, that is, through suprimation of activation of hepatic lipase (lipolysis enzyme that degrades HDL-C).

Estrogens also prevent accumulation of cholesterol and oxidized LDL particles on the arteries wall.

Estrogens alter the transcription of genes coding for several proteins participating in the coagulation system. They affect fibrinogen and factors V, VII, IX X and TFPI and they decrease the levels of anti-thrombinIII, proteinSandPAI-1 [11]. Furthermore, estrogen receptors are expressed on platelets: they influence the migration, the adhesion and the aggregation of these cells and thus increase the thrombotic risk [12]. These effects may offer an explanation for the increased thrombosis events related to the p.o. use of HRT, which were reported in studies such as HERS93, [13] and WHI [14]. In women taking HRT, estrogen dosage, medical history about the inherited hypercoagulable states frequently caused by factor V (Leiden) and prothrombin gene mutations, as well as a history of smoking, are well recognized factors influencing the thrombotic risk associated with HRT [15].

The aim of the paper was to determine the level of follicle stimulating hormone (FSH), estradiol (E2), serum lipids (HDL-CH, LDL-CH / HDL-CH index of arteriosclerosis, LDL-CH, total cholesterol and triglycerides) in women during reproductive life, in perimenopause and postmenopause and to evaluate plasma concentration of coagulation factor VII in the same groups of women.

**Material and methods**

The study included a total of 77 female subjects, divided into 3 groups according the following criteria: the regular (irregular) menstrual cycle; the concentration of serum FSH; the concentration of E2. The control group comprised healthy women (n = 25) with regular menstrual cycle. The hormone level was determined in the late follicular phase (from 10-13 day of the cycle). The second group comprised women in perimenopause (n = 27) with medical history of irregular menstrual cycle, the value of serum FSH being under 25 mIU/ml and the value of E2 above 35 pg/ml. Hormones were determined in the late follicular phase of the cycle. The third group consisted of postmenopausal women (n = 25), with anamnestic data for at least 12 months from the last menstruation, with values of serum FSH above 25 mIU/ml and of E2 under 35 mIU/ml.

Blood samples were collected from an antecubital vein between 8:00 and 9:00 AM, with subjects in the supine position after an overnight fast. For determination of plasma levels of f.VII, blood was anticoagulated with 3.8% trisodium citrate (9:1, vol/vol) and kept on crushed ice until centrifugation. The remaining blood samples were taken without anticoagulant agent (for obtaining serum) and the concentration of hormones FSH and E2 was determined as well as HDL-CH and LDL-CH. After centrifugation at 3000 rpm for 5-10 min., plasma and serum samples were separated and stored at -20°C for further examination.

Hormone concentration was determined with standardized tests based on the radioimmunological method (CIS bio international). Concentration of f. VII was determined with the method of deficient plasma, and serum lipid concentration with the method of fractionation sedimentation according to the specific weight (MERCK). Measurement was done on spectrophotometer at wavelength - 500 nm.

Data were entered into a data-base and were statistically analyzed, p < 0.05 being considered as statistically significant difference.
Results
The investigation has shown that there were differences in the levels of hormones, factor VII and lipid parameters among the three groups of examined women. Table 1 presents mean values of serum concentration of hormones FSH and estradiol (E2) as well as factor VII in the three examined groups of women.

Table 1. Concentration of reproductive hormones and factor VII in women during menopause (n=77)

<table>
<thead>
<tr>
<th></th>
<th>Control group n=25</th>
<th>Perimenopause n=27</th>
<th>Postmenopause n=25</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>FSH mIU/ml</td>
<td>4.6 ± 3.6</td>
<td>17.2 ± 10.5</td>
<td>70.5 ± 21.1</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>E2 pg/ml</td>
<td>101.9 ± 58.6</td>
<td>36.2 ± 23.2</td>
<td>10.2 ± 8.6</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Factor VII %</td>
<td>80.4 ± 8.5</td>
<td>87.2 ± 16.9</td>
<td>110.1 ± 12.9</td>
<td>&lt; 0.05</td>
</tr>
</tbody>
</table>

Each values is an arithmetic mean value ± standard deviation of 77 single examinations

It is evident that there is a statistically significant (p<0.001) increase of serum concentration of FSH hormone, statistically significant (p<0.001) decrease of serum concentration of E2 and increase (p<0.05) of the concentration of coagulation factor VII in different phases of the reproductive life of the women.

Mean values of concentrations of serum lipids (HDL-CH, LDL-CH) in women during different phases of the reproductive life are presented in Table 2.

Table 2. Serum lipids in women according menopausal status (n=77)

<table>
<thead>
<tr>
<th></th>
<th>Control group n=25</th>
<th>Perimenopause n=27</th>
<th>Postmenopause n=25</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>HDL-CH mmol/l</td>
<td>2.2 ± 0.5</td>
<td>1.4 ± 0.4</td>
<td>1.2 ± 0.2</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>LDL-CH mmol/l</td>
<td>2.4 ± 0.9</td>
<td>3.2 ± 12</td>
<td>4.6 ± 1.3</td>
<td>&lt; 0.001</td>
</tr>
</tbody>
</table>

Each values is an arithmetic mean value ± standard deviation of 77 single examinations

A statistically significant decrease (p< 0.001) was evident in the serum concentration of HDL-CH and statistically significant increase (p < 0.001) in the level of LDL-CH in perimenopausal and menopausal women when compared with those in the control group.

Discussion
The increase of factor VII in postmenopausal women could be explained with changes that happen in the endothelium and are a result of absence of estrogens and their antioxidative effect, which under circumstances of impaired lipid profile (increase of LDL-C and its oxidation),
creates initial conditions for appearance of atherogenic lesions. This most probably leads to activation of the coagulation process where factor VII plays a key role.

Vast number of literature data report on the changes in the hemostatic system with advancing age and these are in direct relation with estradiol status. Fibrinogen level and factor VII activity [16] markedly increase in the period of postmenopause.

In our study we have shown that there was also a profound decrease of HDL-C concentration and increase of LDL-CH in both perimenopausal and postmenopausal women as compared with the controls.

Protective effect of estrogens in the development of atherosclerosis has been proved in numerous experimental and clinical studies [17].

Lipid profile in females and males of comparable age has been examined and it has been proved that the level of LDL-CH, apo B and triglycerides is higher in men than in women during fertile period. These parameters increase in menopausal women [18]. Increase of LDL-CH and decrease of HDL-CH is evident risk factor for coronary heart disease. Women in fertile period have significantly lower values of LDL-C, apo B and all lipoproteins containing apo B, but they also have higher level of HDL-C and apo A-1, a major HDL lipoprotein, as well as higher percentage ratio HDL2/HDL3 when compared to women in menopause [19]. Disorders of lipid profile in perimenopausal and postmenopausal women increase the risk of coronary heart diseases. Application of estrogen substitutional therapy significantly decreases this risk. Namely, level of LDL-CH is significantly decreased in postmenopausal women after administration of estrogen substitution, whereas HDL-CH and apo A-1 are significantly increased [20].

**Conclusion**

Due to our results we can conclude that decrease in estradiol level and associated decrease in HDL-CH, and increase in LDL-CH, PAI-1 Ag and factor VII of coagulation seen in perimenopausal and postmenopausal women may be responsible for the increased risk of atherosclerotic complications and cardiovascular diseases in women during menopause.

**References**