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SERUM LEPTIN IN THE DEVELOPMENT OF INSULIN RESISTANCE AND OTHER DISORDERS IN THE METABOLIC SYNDROME

Radka Lichnovská, Simona Gwozdziewiczová, Rudolf Chlup, Jiří Hřebíček†

Institute of Physiology, Faculty of Medicine, Palacký University, 775 15 Olomouc, Czech Republic
e-mail: lichnov@tunw.upol.cz

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The metabolic syndrome mostly represented by obesity and hyperinsulinaemia connected with insulin resistance, presents the main mechanism in the pathogenesis of cardiovascular disease. The aim of this study was to analyze the interrelations between several metabolic variables (including leptin) and factors related to insulin resistance in groups of both normal and non-diabetic hyperlipemic postmenopausal women and men of appropriate age, and to attempt to elucidate the gender differences. Two groups of patients (20 men, 20 women) with hypertriglyceridemia were compared with 30 individuals (10 men, 20 women) with normal serum triacylglycerols. Fasting serum leptin concentration, lipid parameters (triacylglycerols, HDL cholesterol, LDL cholesterol) and BMI were measured and compared with changes in insulin parameters influencing insulin resistance (HOMA IR, insulin, intact proinsulin, C-peptide). Statistical analysis was performed using SAS/STAT software including unpaired Student's t-test, Kolmogorov-Smirnov's test, Spearman's rank-order correlation and multiple regression analysis. In men, the insulin sensitivity correlates with leptin only. In women insulin sensitivity is markedly influenced by a complex of factors: leptin and lipid parameters. Increased insulin resistance in men is followed mainly by the increased correlations between leptin, HOMA IR and insulin parameters. In women correlations between leptin, HOMA IR and insulin parameters were smaller, but the inverse correlation with HDL cholesterol was stronger. In postmenopausal women and also in men, serum leptin concentration contributes to insulin resistance. However in women the effect of increase in serum triacylglycerols in contribution of insulin resistance seems to be more dominant.

INTRODUCTION

The metabolic syndrome (Reaven's syndrome) mostly represented by obesity and hyperinsulinaemia connected with insulin resistance, presents the main mechanism in the pathogenesis of cardiovascular disease. The prevalence of this metabolic disorder constantly increases in connection with the pandemia of obesity and insulin resistance. Noted components appear in younger individuals even in children[24, 30].

More than every third adult human suffers from this syndrome. Single factors of the metabolic syndrome hang together and influence each other. Therefore the effect does not add up, but instead multiplies and increases the risk of cardiovascular morbidity and mortality[13, 18, 24, 25].

Traditionally, adipose tissue was considered as a passive reservoir of energy in the form of triacylglycerol stores and as a place for the conversion of androgens to estrogens. In recent years, a number of papers have stated, that adipocytes produce many hormones and adipocytokines such leptin, TNFα, resistin, adiponectin, adipsin etc. These influence energy homeostasis, balance of fat stores and make possible adaptation of the organism to various situations (starvation, stress, infection and period of increased energy demand)[9, 21].

Leptin discovered in 1994 is a multiple-function cytokine involved in the regulation of food intake, energy metabolism and sacharide and lipid metabolism. Impaired regulation of food intake in consequence of leptin resistance is presented in connection with the aetiopathogenesis of obesity and insulin resistance, but its role in development of these diseases is not still clear.

AIMS

The aim of this study was to estimate serum leptin concentration in groups of both normal and non-diabetic hyperlipemic postmenopausal women and men of appropriate age, to analyze the interrelations between several metabolic variables (including leptin) and factors related to insulin resistance and to attempt to elucidate the gender differences.

MATERIAL AND METHODS

1) Persons in the study

The study was carried out on 70 persons at the Metabolic Centre of the Hospital in Šternberk, Czech Republic.

From these, 40 patients (20 men and 20 women) with typical dyslipidemia were selected. 20 men with plasma triacylglycerol concentrations exceeding 1.5 mmol/l and with HDL cholesterol concentration under 1.1 mmol/l were denominated as test group. The test group of postmenopausal women consisted of 20 individuals with plasma triacylglycerol concentrations exceeding 1.7 mmol/l and with HDL cholesterol concentration under 1.4 mmol/l. The average age of male was 58.5 ± 10.4 y, and of female 59.9 ± 13.0 y, respectively. These criteria are typical for the early stage of the metabolic syndrome. No subject had serious complications. Two other groups (10 men and 20 women) with approximately normal serum values of these variables were taken as controls. The average age for male was 60.3 ± 11.0 y, and for female 59.9 ± 13.0 y, respectively. None of the persons in either control or test groups had clinically apparent diabetes mellitus, but some of the hyperlipidemic patients showed impaired glucose tolerance (6.1–7.0 mmol/l) or impaired fasting glucose (6.1–7.8 mmol/l). All hyperlipidemic patients were treated with antihyperlipidemic drugs, especially fibrates. Some of these patients were treated with antihypertensive therapy. No signs of major clinical or laboratory symptoms of other diseases were present in any group. Blood samples were obtained in the morning by a venous puncture after overnight fasting. After clotting the serum was separated and stored at – 20° until used. Informed consent was obtained from all probands.

2) Biochemical methods

Serum leptin concentrations were measured by a sandwich ELISA test kit (Human Leptin ELISA, BioVendor Laboratory Medicine, Inc, Czech Republic). Its sensitivity limit was 0.2 ng/ml, intraassay CV 6.1 % at the level of 7.5 ng/m, inter-assay CV 8.5 % at the level of 4.8 ng/ml. Tetramethylbenzidine was used as a substrate; quality controls were human based. Several other hormones and peptides were estimated by routine immunochemical tests: insulin, C-peptide (IMMULITE, Diagnostic Products Corporation, Los Angeles, CA, U.S.A.), proinsulin intact (DAKO, Denmark). Serum concentration of glucose, total cholesterol, triacylglycerols, HDL-cholesterol, LDL-cholesterol and uric acid were measured on ILAB-600 biochemical analysator (Instrumentation Laboratory, Lexington, Ma, U.S.A.) using BioVendor sets. All samples were processed and examined according to the principles of good laboratory practice and under permanent intralaboratory and external quality control.

3) Indexes

a) Body mass index (BMI) = body mass (kg) /height (m)

b) The homeostatic index of insulin resistance (HOMA IR) was calculated according to the homeostasis model of assessment[10, 17] as follows: **fasting insulin (μU/ml). fasting glukose (mmol/l) / 22.5**

4) Statistics

Statistical analysis was performed using the Version 6 SAS/STAT software (SAS Institute, Inc., Cary, NC, U.S.A.). The statistical significance of differences between the means in the test and control groups were evaluated using the unpaired Student's t-tests in the case of normal distribution of data sets, and using the Kolmogorov-Smirnov's test when at least in one of the data sets the normal distribution was excluded. Spearman's rank-order correlation was used for correlation analysis. $p < 0.05$ was considered to be statistical significance. Multiple regression analysis was performed using parameters typical for insulin resistance (HOMA IR, intact proinsulin and C-peptide) as dependent variables, and other metabolic factors (lipid parameters, BMI and leptin) as independent variables. The so called step-down regression model was used to select dominant independent variables. Various four-member groups of independent (explanatory) variables were used for the analysis and the non-zero intercept was taken into account. The independent variables were then dropped, one at a time: at each stage one variable making the least contribution to the dependent variable (i.e. that showed the least p-value in the test of the regression coefficient being zero) was excluded. The coefficient of determination R^2 that can be viewed as a percentage explaining the total variance was simultaneously monitored. A great drop of R^2 after excluding some independent variable enabled to select those independent variables that could be thought to be the most important determinants of the dependent variable.

RESULTS

Table 1A and 1B demonstrate evaluation of the differences between mean parameters in control and test groups of men and women, respectively. Big differences between test and control groups in HOMA IR, serum concentrations of uric acid, triacylglycerols and HDL cholesterol were found. The average concentration of leptin in test group of men is twice high and statistic significant as compared with control group of men. In case of women the situation is similar; however, the results are rather different: Changes in concentrations of insulin and lipid parameters were found, however without any significant increase in leptin, C-peptide and uric acid.

In Table 2A and 2B the results of Spearman's correlations between parameters of insulin resistance, various metabolic parameters and leptin are presented. In both groups of men (see Table 2A and 2B), positive significant correlations between leptin, HOMA IR and insulin, between serum triacylglycerols and intact proinsulin were found. According to the correlation analysis the increase in insulin resistance in men is followed mainly by increase in correlations between leptin, HOMA IR and insulin parameters typical for insulin resistance (insulin, proinsulin and mainly C-peptide).

In Table 3A and 3B the results of Spearman's correlations between similar parameters in both groups of women are presented. In the control group (see Table 3A) we found positive correlations between leptin, insulin and HOMA IR as well as in men. On the other hand lep-

Table 1A and 1B. Comparison of the control and the test groups of men and women respectively˝

1A: MEN		Control group Average	SD	Test group Average	SD	P
Age	(years)	60.30	11.07	58.65	10.47	0.69
BMI		25.91	3.58	28.51	2.60	0.0307
Uric acid	(μmol/l)	270.50	65.40	379.95	84.17	0.0029
Glycaemia	(mmol/l)	5.31	0.53	6.11	0.95	0.0291_{KS}
Cholesterol	(mmol/l)	5.07	1.06	6.62	0.82	0.0001
Triacylglycerols	(mmol/l)	1.11	0.44	3.52	1.38	0.0001
HDL-cholesterol	(mmol/l)	1.43	0.39	0.96	0.24	0.0003
LDL-cholesterol	(mmol/l)	3.08	0.83	4.39	1.06	0.0047
Insulin	(mIU/l)	6.96	2.80	11.43	4.50	0.0126_{KS}
HOMA IR		1.69	0.81	3.14	1.46	0.0017_{KS}
Proinsulin	(pmol/l)	2.68	1.15	5.51	2.75	0.0126
C-peptide	(nmol/l)	0.66	0.31	1.11	0.39	0.0148
Leptin	(ng/ml)	3.07	3.83	6.21	3.78	0.0126_{KS}

1B: WOMEN		Control group Average	SD	Test group Average	SD	P
Age	(years)	56.95	13.03	62	7.36	0.15
BMI		25.36	3.72	26.73	3.59	0.24
Uric acid	(μmol/l)	236.15	67.06	270.80	58.93	0.13
Glycaemia	(mmol/l)	5.26	0.42	5.85	0.70	**0.0027**
Cholesterol	(mmol/l)	5.15	0.70	6.92	1.01	**0.0001**
Triacylglycerols	(mmol/l)	1.42	0.47	2.72	0.92	0.0001_{KS}
HDL-cholesterol	(mmol/l)	1.50	0.21	1.21	0.21	**0.0001**
LDL-cholesterol	(mmol/l)	3.21	0.71	4.74	1.02	**0.0001**
Insulin	(mIU/l)	7.33	3.80	10.27	3.52	0.0348_{KS}
HOMA IR		1.73	0.97	2.69	1.04	0.0015_{KS}
Proinsulin	(pmol/l)	2.51	1.76	4.49	3.11	0.0348_{KS}
C-peptide	(nmol/l)	0.748	0.34	0.92	0.35	0.22_{KS}
Leptin	(ng/ml)	16.06	13.49	14.79	5.89	0.17_{KS}

The statistical significance of differences between the means in the test and the control groups were evaluated using the unpaired Student's t-test in the case of normal distribution of data sets, and using the Kolmogorov-Smirnov's (KS) test when at least in one of the data sets the normal distribution was excluded ($p < 0.05$).

tin positively correlated with proinsulin, C-peptide, LDL cholesterol and negatively with HDL cholesterol. We also found significant positive correlation between LDL cholesterol, insulin and proinsulin and negative correlations between HDL cholesterol, insulin and HOMA IR. In women without signs of increased insulin resistance the sensitivity of insulin is markedly influenced with a complex of factors: leptin and lipid parameters (LDL cholesterol and decreased HDL cholesterol). However in men, the sensitivity of insulin correlates only with leptin.

In the test group of women (see Table 3B), Spearman's correlations between leptin, HOMA IR and insulin parameters were smaller. Inverse correlations between HDL cholesterol and these parameters increased.

Table 4A, 4B and 4C show results of multiple regression analysis, when data from both control and test groups of men were judged together. HOMA IR is mostly influenced by concentration of serum leptin. Serum proinsulin is mostly influenced by decreased HDL cholesterol together with increase in BMI. The chosen constellation of independent variables mostly influences the concentration of serum C-peptide. Leptin especially plays a dominant role together with serum triacylglycerols.

Table 2A and 2B. Spearman's correlation coefficient between parameters of insulin resistance, various metabolic parameters and leptin in men (control and test groups)

2A: Control group	BMI	TAG	HDL	LDL	Leptin
Insulin	S_k = 0.188	S_k = 0.60	S_k = 0.006	S_k = 0.428	**S_k = 0.652 p = 0.041**
HOMA IR	S_k = 0.248	S_k = 0.515	S_k = 0.006	S_k = 0.428	**S_k = 0.658 p = 0.038**
Proinsulin	S_k = – 0.479	**S_k = 0.733 p = 0.02**	S_k = – 0.479	S_k = – 0.047	S_k = 0.353
C-peptide	S_k = 0.257	S_k = 0.143	S_k = 0.428	S_k = 0.400	S_k = 0.145
Leptin	S_k = 0.317	S_k = 0.006	S_k = 0.140	S_k = 0.012	

2B: Test group	BMI	TAG	HDL	LDL	Leptin
Insulin	S_k = 0.166	S_l = – 0.257	S_k = – 0.237	S_k = – 0.001	**S_k = 0.449 p = 0.046**
HOMA IR	S_k = 0.358	S_k = – 0.278	S_k = – 0.180	S_k = – 0.057	**S_k = 0.574 p = 0.008**
Proinsulin	S_k = 0.406	S_k = 0.129	S_k = – 0.362	**S_k = – 0.543 p = 0.013**	S_k = 0.407
C-peptide	**S_k = 0.562 p = 0.0098**	S_k = 0.249	S_k = – 0.432	S_k = – 0.192	**S_k = 0.803 p = 0.0001**
Leptin	**S_k = 0.765 p = 0.0001**	S_k = 0.057	S_k = – 0.137	S_k = – 0.084	

Statistically significant correlations ($p < 0.05$).

Table 5A, 5B and 5C show the results of multiple regression analysis, when data from both control and test groups of women were judged together. In the case of HOMA IR high significant values of R^2 were achieved in combination of these independent variables. Serum triacylglycerols play a dominant role together with the serum leptin. Serum proinsulin is mostly determined by the concentration of serum triacylglycerols together with the decreased HDL cholesterol. Serum C-peptide is mostly influenced by the concentration of the serum triacylglycerols together with leptin.

These correlations demonstrate some physiological mechanisms, which participate in regulation of insulin sensitivity, however some differences in men and women respectively. We have decided to investigate these differences in pathogenesis of insulin resistance between men and women by multiple regression analysis using parameters typical for insulin resistance (HOMA IR, intact proinsulin and C-peptide) as dependent variables, and metabolic factors involving leptin, lipid parameters and BMI as independent variables. Data from both control and test groups of each gender were judged together.

DISCUSSION

Insulin resistance and changes in lipid parameters are typical for early signs of the metabolic syndrome. Hypertriacylglycerolaemia together with decreased of HDL cholesterol were pivotal criteria in the selection of the groups. We were compared the results of control probands with the results of hyperlipidemic persons in test groups. All parameters related to insulin resistance (plasma concentration of glucose, insulin, proinsulin, C-peptide, triacylglycerols, HDL cholesterol, uric acid) were typically changed in the test group of men. Furthermore, the concentrations of LDL cholesterol and total cholesterol are increased too (see Table 1).

The results from the test group of women are rather different. We found no increase in uric acid, C-peptide in serum. However, other parameters are typical for manifestations of the metabolic syndrome: significant increase in fasting glycaemia, triacylglycerols, fasting insulin and proinsulin, together with increases in total cholesterol, LDL cholesterol and decrease in HDL cholesterol.

A significant increase in BMI only in the test group of men was presented. However the increase in absolute values of BMI is not so pronounced (from 25.9 to 28.5) and it ranges in the overweight category but not in obesity. The values of BMI in both groups of women are in

Table 3A and 3B. Spearman's correlation coefficient between parameters of insulin resistance, various metabolic parameters and leptin in women (control and test group)

3A: Control group	BMI	TAG	HDL	LDL	Leptin
Insulin	S_k = 0.399	S_k = 0.169	**S_k = – 0.498 p = 0.025**	**S_k = 0.523 p = 0.037**	**S_k = 0.813 p = 0.0001**
HOMA IR	S_k = 0.354	S_k = 0.157	**S_k = – 0.499 p = 0.025**	**S_k = 0.393**	**S_k = 0.826 p = 0.0001**
Proinsulin	S_k = 0.163	S_k = 0.009	S_k = – 0.243	**S_k = 0.563 p = 0.023**	**S_k = 0.471 p = 0.0358**
C-peptide	S_k = 0.176	S_k = 0.332	S_k = – 0.356	S_k = 0.027	**S_k = 0.724 p = 0.0117**
Leptin	S_k = 0.366	S_k = 0.296	**S_k = – 0.512 p = 0.0208**	**S_k = 0.505 p = 0.046**	

3B: Test group	BMI	TAG	HDL	LDL	Leptin
Insulin	S_k = 0.261	S_k = 0.292	**S_k = – 0.520 p = 0.018**	S_k = – 0.046	**S_k = 0.494 p = 0.026**
HOMA IR	S_k = 0.308	S_k = 0.334	**S_k = – 0.554 p = 0.011**	S_k = – 0.111	**S_k = 0.471 p = 0.036**
Proinsulin	S_k = – 0.052	S_k = 0.391	**S_k = – 0.516 p = 0.019**	S_k = – 0.207	S_k = 0.142
C-peptide	S_k = 0.424	S_k = 0.230	S_k = – 0.473	S_k = – 0.314	**S_k = 0.726 p = 0.003**
Leptin	**S_k = 0.509 p = 0.021**	S_k = – 0.110	S_k = – 0.407	S_k = – 0.311	

Statistically significant correlations ($p < 0.05$).

the overweight category too (about 25–26). We conclude that these gender differences of BMI are not substantial; however these could influence some parameters such as adiposity.

In compliance with other studies the serum leptin concentration in the control group of women is fivefold higher as compared with men. In the test group of women more than fivefold concentration of leptin does not increase without any connection with the changes in BMI. In the test group of men the serum leptin concentration is significantly higher as compared with controls. We could assign these changes to the differences in the raise of BMI in men, but absolute differences in BMI are so small.

The serum leptin concentration significantly correlates with HOMA IR which reflects the degree of insulin resistance and with the concentration of serum insulin in both control and test groups of men and women respectively. We can place our results along the large group of studies, which declare an important role of leptin in pathogenesis of insulin resistance[1, 2, 4, 6, 8, 11, 15, 20, 28, 29, 31].

Although the concentration of serum leptin in the control group of women is manifestly higher than in the control group of men, the average index of insulin resistance HOMA IR and the average concentration of serum insulin in both control groups are not very different. However, the correlation between leptin, HOMA IR and insulin is very important ($p < 0.0001$). Leptin takes part in the influence of insulin resistance, but in case of women it is necessary substantially higher concentrations of leptin as compared with men. In control group of women HOMA IR, concentrations of serum leptin and insulin significantly negatively correlate with HDL cholesterol and positively with LDL cholesterol as compared with men (see Table 2 and 3). Leptin is also importantly correlated with C-peptide. In women without increased insulin resistance the sensitivity to insulin bears on a complex of factors including leptin, HDL and LDL cholesterol. In the case of men this sensitivity to insulin correlates only with leptin.

In women increased insulin resistance with manifestations of the metabolic syndrome leads to quantitative changes: the weakening of correlation between leptin, HOMA IR and insulin parameters and simultaneously the emphasis of the influence of HDL cholesterol on insulin parameters. In men the correlation between leptin, HOMA IR and especially between C-peptide is emphasised. Only one qualitative change was found in men and women, respectively: the correlation between leptin and BMI. In men another correlation was found: the correlation between BMI and C-peptide (see Table 2 and 3).

Generally, the increase of insulin resistance in men is mostly followed by increase in correlations between leptin, HOMA IR and insulin parameters typical pro insulin re-

Table 4A, 4B and 4C. Multiple regression analysis of data from men (controls and tests)

		Intercept	BMI	Leptin	HDL	TAG	R^2
4A: HOMA IR	Par.	0.5423	0.0983	0.1300	– 1.0709	– 0.0320	**0.3947**
	T = 0	0.8104	0.3112	0.1009	0.1902	0.8511	
	Par.	0.5249	0.0923	0.1321	– 0.9931		**0.3938**
	T = 0	0.8127	0.3037	0.0865	0.1495		
	Par.	2.6226		0.1795	– 0.8047		**0.3681**
	T = 0	0.0072		0.0051	0.2231		
	Par.	1.5886		0.2064			**0.3317**
	T = 0	0.0001		0.0009			

		Intercept	BMI	Leptin	HDL	TAG	R^2
4B: Proinsulin	Par.	2.5975	0.1473	0.1247	– 2.9601	0.2011	**0.4070**
	T = 0	0.5346	0.4093	0.3841	0.0544	0.5245	
	Par.	2.7071	0.1851	0.1118	– 3.4484		**0.3971**
	T = 0	0.5124	0.2679	0.4245	0.0101		
	Par.	1.5474	0.2659		– 3.8953		**0.3819**
	T = 0	0.6865	0.0488		0.0016		
	Par.	8.9041			– 3.9003		**0.2844**
	T = 0	0.0001			0.0024		

		Intercept	BMI	Leptin	HDL	TAG	R^2
4C: C-peptid	Par.	– 0.6664	0.0484	0.0454	– 0.1261	0.0677	**0.6764**
	T = 0	0.2405	0.0524	0.0148	0.4962	0.0942	
	Par.	– 0.7078	0.0430	0.0501		0.0793	**0.6690**
	T = 0	0.2050	0.0628	0.0039		0.0310	
	Par.	0.3252		0.0669		0.1002	**0.6112**
	T = 0	0.0227		0.0001		0.0083	
	Par.	0.5982		0.0716			**0.4699**
	T = 0	0.0001		0.0001			

T = 0 – p-value in the test of the regression coefficient being zero, R^2 – coefficient of determination, expressing the degree of influence of dependent variable by independent variables. Dominant and statistically important regression coefficients are designated.

sistance (insulin, proinsulin, especially C-peptide). In the case of women with insulin resistance, these correlations are also expressed (especially correlation between leptin and C-peptide), but correlations between HDL cholesterol and these parameters are emphasised.

Kennedy et al.[14] reports that serum leptin increases in proportion to progression of obesity. It correlates with BMI and with adiposity in both genders, especially in women. In every stage of obesity, the concentration of serum leptin was much higher in women than in men. Authors characterise this fact as "state of relative leptin resistance" in women corresponding with the different physiognomy of the genders. However, our results suggest that serum leptin concentrations in women can rise independently of the increase of BMI in connection with the increase of insulin resistance. Increased BMI in our test group of men was not so prominent for explanation of the great increase of serum leptin concentration.

On the other hand Clapham JC et al.[3] found, that serum leptin in middle age women correlated with BMI and adiposity especially in non obese women and correlated with the sensitivity of insulin neither in non obese women nor in obese individuals. Coullard C et al.[5] found substantially higher values of serum leptin in women and the correlation between leptin and body fat (determined by weighing under water and using CT) in both genders. Gender differences in serum leptin concentration were present even when comparing men and women with the same body fat mass. In both gender higher serum leptin concentrations were associated with higher fasting serum insulin. In contrast to men higher levels of serum leptin in women was independent of the total body fat mass. It is evident, that correlation between leptin and insulin resistance in women evidently need not be dependent on raising obesity in women.

The large population study[19] shows, that BMI is the most important determinant of serum leptin in women. This finding does not comply with our results. This study[19] also confirmed positive correlation between serum leptin and insulin and found relationships between serum leptin and triacylglycerols independent of gender, BMI, waist ratio and serum insulin. Hattori A et al.[12] found,

Table 5A, 5B and 5C. Multiple regression analysis of data from women (controls and tests)

		Intercept	BMI	Leptin	HDL	TAG	R^2
5A: HOMA IR	Par.	1.9074	0.0124	0.0385	– 1.1471	0.4578	**0.6148**
	T = 0	0.1844	0.7359	0.0070	0.0766	0.0056	
	Par.	2.1774		0.0402	– 1.1473	0.4704	**0.6135**
	T = 0	0.0662		0.0023	0.0728	0.0032	
	Par.	0.1308		0.0495		0.6380	**0.5768**
	T = 0	0.6822		0.0001		0.0001	
	Par.	0.7954				0.6856	**0.3664**
	T = 0	0.0223				0.0001	

		Intercept	BMI	Leptin	HDL	TAG	R^2
5B: Proinsulin lin	Par.	5.5486	0.0259	0.0098	– 3.7693	1.0848	**0.4788**
	T = 0	0.1725	0.8029	0.7981	0.0411	0.0183	
	Par.	6.1133		0.0134	– 3.7698	1.1111	**0.4779**
	T = 0	0.0679		0.7005	0.0384	0.0120	
	Par.	6.7597			– 4.0480	1.0812	**0.4757**
	T = 0	0.0190			0.0147	0.0118	
	Par.	– 0.0225				1.7038	**0.3829**
	T = 0	0.9777				0.0001	

		Intercept	BMI	Leptin	HDL	TAG	R^2
5C: C-peptid	Par.	1.0638	– 0.0209	0.0216	– 0.2806	0.1469	**0.5206**
	T = 0	0.1710	0.2839	0.0063	0.4488	0.0743	
	Par.	0.5810	0.0213	0.0236		0.1882	**0.5063**
	T = 0	0.1703	0.2696	0.0016		0.0031	
	Par.	0.1555		0.0197		0.1573	**0.4760**
	T = 0	0.3537		0.0018		0.0045	
	Par.	0.5412		0.0173			0.2370
	T = 0	0.0004		0.0136			

T = 0 – p-value in the test of the regression coefficient being zero, R^2 – coefficient of determination, expressing the degree of influence of dependent variable by independent variables. Dominant and statistically important regression coefficients are designated.

that leptin correlated with HOMA IR and BMI in healthy and diabetic men, but in the same groups of women only with BMI. Correlations between HOMA IR and serum leptin were dependent on BMI. These findings do not correspond with our results. In another study highly significant correlations were found between body mass, body fat mass and serum leptin in middle-age men and pre- and postmenopausal women[23]. The main determinant of serum leptin and insulin concentrations is only body mass in men, body fat mass in premenopausal women and body mass in postmenopausal women. The authors do not define state of insulin resistance in case of their test persons; therefore comparison with our results is not possible.

Various proportion of body fat mass may explain the dependence of leptin on gender, age and race[16]. Leptin more correlates with proportion of body fat than with total body fat mass or with BMI[22]. In women leptin secretion from subcutaneous adipocytes in abdominal region is about three times higher than from intraabdominal (omental) adipose tissue. Subcutaneous adipocytes are the main source of serum leptin[27].

In men and women with impaired glucose tolerance only correlations between level of obesity and serum leptin concentration were found[7,26]. The authors conclude that gender and other differences in leptin concentration almost entirely bear on regional distributions of adipose tissue.

CONCLUSIONS

The significant role of serum leptin in determination of insulin resistance in both elderly men and postmenopausal women of equal age was confirmed. In men leptin is a dominant factor and together with the decrease in HDL cholesterol it can influence the degree of insulin resistance and together with the increase in serum triacylglycerols it can participate in release of insulin from endogenous pancreas. In postmenopausal women leptin also plays an important role in this process, but the effect of increase in serum triacylglycerols in contribution of insulin resistance seems to be more dominant. Further

studies are necessary to assess the roles of individual factors in the development of insulin resistance.

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