Circulating Chemerin is Associated with Subclinical Atherosclerosis in Obesity

VIVIANA AURSULESEI[1]*, ECATERINA ANISIE[2], ANA MARIA ALECSA[3], MARIA MAGDALENA LEON CONSTANTIN[1], RAZAN AL NAMAT[1]

[1] Grigore T. Popa University of Medicine and Pharmacy, Faculty of Medicine, 1st Medical Department, 16 Universitatii Str., 700115, Iasi, Romania

[2] Sf. Spiridon Emergency Districtual Clinical Hospital, Immunology and Genetics Lab, 1 Independentei Sq., Iasi, 700111, Romania

[3] Sf. Spiridon Emergency Districtual Clinical Hospital, Department of Radiology, 1 Independentei Sq., 700111, Iasi, Romania

Chemerin is a newly discovered adipokine with controversial role in obesity. The relationship between circulating chemerin and noninvasive biomarkers of subclinical atherosclerosis is far from being elucidated. Our study concluded that chemerin could be an independent predictor for arterial stiffness, but not for mean carotidian intima media thickness and epicardial fat thickness. Most probably, the effects of chemerin are modulated by concurrent metabolic factors and mechanisms involved in early atherosclerosis.

Keywords: chemerin, obesity, subclinical atherosclerosis

Visceral adipose tissue is a wide source of biologically active molecules termed adipokines. The pattern of adipokines secretion directly influences the development of comorbidities related to obesity [1]. Growing evidence suggests that in obesity adipokines generate oxidative stress, inflammation, insulin resistance (IR), macrophage activation, thus mediating the atherosclerosis (ATS) [2-4]. Some studies link circulating adipokines to vascular structural changes involved in early vascular aging [5]. It has been reported that in obesity or diabetes mellitus both local and circulating adipokines may be independently associated with markers of subclinical ATS, such as pulse wave velocity, central blood pressure or carotid intima-media thickness [6, 7]. However, the knowledge of this relationship is limited for obesity.

A potential candidate for study is chemerin, a newly discovered adipokine with controversial reported role in obesity [8]. Chemerin is involved in adipogenesis, adipocyte metabolism and the pathogenesis of IR [9-12]. Clinical data that refers to its relationship with obesity and metabolic syndrome are also conflicting [13-16]. A recent meta-analysis confirms that the serum levels correlate with body mass index (BMI) and IR, so it could play an important pathophysiological role [17]. Although some signals link chemerin to early atherosclerotic plaque development, morphology and progression [18], there are more controversial assumptions rather than consistent information [19-23]. The relationship between chemerin and adiponectin as a possible mechanism involved in ATS also requires further research [24, 25].

Thus, our study aimed to investigate the relationship of chemerin with noninvasive validated parameters of subclinical ATS in obese subjects.

Experimental part
Material and methods

Fifty participants including 25 obese patients (BMI $\geq$ 35 kg/m^2) who were admitted for bariatric surgery and 25 age- and gender matched non-obese control patients (BMI < 30 kg/m^2) were enrolled in a cross-sectional study. Patients with concurrent enrollment in another study, with documented cardiovascular or metabolic disease, with comorbidities generating inflammation or factors that could influence measurement of the arterial stiffness and the parameters of inflammation (smoking, treated hypertension, vasodilators, lipid-lowering agents, estrogens, anti-inflammatory drugs, antioxidants) were excluded. All included subjects had no criteria for defining metabolic syndrome, i.e. the presence of any 3 of 5 risk factors according to the 2009 Joint Interim Statement [31]. The protocol has been explained in detail and the informed consent was signed before enrollment. The study protocol was approved by the University- and the Hospital Local Ethics Committees. Anthropometric measurements included BMI (kg/m^2), waist circumference, waist to hip circumference ratio (WHR) and index of central obesity (waist circumference to height ratio). Venous blood samples collected for the assessment of biochemical parameters linked to the cardiovascular risk (plasma cholesterol, HDL-cholesterol, LDL-cholesterol, triglycerides, fasting plasma glucose, uric acid, creatinine, high-sensitivity C-reactive protein – hs-CRP) were processed within two hours. The venous blood samples collected for assessment of chemerin and adiponectin were stored at -20°C and processed after completing the enrollment of patients. Serum chemerin and adiponectin were measured quantitatively by specific Human ELISA (enzyme-linked immunosorbent assay) kits (ab155430, ab99968, respectively) supplied by Abcam Cambridge, U.K., for research use only. Chemerin/adiponectin ratio was calculated and the relation with subclinical ATS was also studied. Chemerin (known as retinoic acid receptor responder protein 2 - RARRES2, tazarotene-induced gene 2 protein - TIG2), in humans encoded by the RARRES2 gene, is a 14 kDa protein and consists of 163 aminoacids. It is secreted as inactive prochemerin, subsequently activated by different proteases. Chemerin is a multifunctional protein with chemotactic properties for leukocytes that express receptor CMKLR1 (or ChemR23) [8]. It was discovered as an adipokine when the secretion by adipocytes, the high mRNA expression with CMKLR1 receptor in white adipose tissue [32], the role in adipocyte differentiation and glucose uptake were demonstrated. Adiponectin (or adipocyte complement-related protein of 30 kDa) is exclusively secreted from adipose tissue as a monomer. The biological functions are influenced by the

email: aursuleseiv@yahoo.com; Phone: (+40)751113451

assembly in homooligomers of various molecular weights. The high-molecular weight form is the most active and is related to a lower risk of diabetes [33].

Subclinical atherosclerosis was assessed noninvasively by several validated parameters. Arterial stiffness was determined according to 2012 Expert Consensus Document [34]. A validated Tensiomed™-Arteriograph TL2 device (TensioMed Ltd., Budapest, Hungary) was used. The studied parameters were aortic pulse wave velocity (PWV), brachial and aortic augmentation index (Aix), including the adjusted value to a heart rate of 75 beats per minute – Aix 75, systolic aortic blood pressure (SBPao), aortic pulse pressure (PPao) and subendocardial viability ratio (SEVR) also known as Buckberg index. Carotidian intima media thickness (IMTc) was evaluated by high-resolution B-mode system (Aloka - ProSound SSD-3500SX, Aloka Co., LTD., Tokyo, Japan), equipped with a 7-12 MHz linear array transducer. Mean IMTc values were measured from the far walls of the right and left common carotid artery (mean-mean) [35]. Epicardial fat thickness, functionally considered a visceral fat deposit, was measured on the free wall of the right ventricle from the parasternal long axis and short axis views at end-systole [36].

Statistical analysis

Data analysis was performed using IBM SPSS Statistics Version 22.0. The variables were described using mean values ± standard deviation (SD). Independent two-sample test was used to study the differences between the obese and non-obese samples. A natural logarithmic transformation was performed for the variables without a normal distribution. Thus, Pearson coefficient was used to analyze the linear correlation between serum chemerin and the studied variables. Kendall tau coefficient was used to determine the same correlations for the whole sample. Multiple linear regression models were created to analyze the risk factors that determine arterial stiffness in obese sample. The most important condition when creating these models was that serum chemerin was a significant variable in the model.

Results and discussions

The demographic, clinical and biochemical characteristics are presented in table 1. The mean age of the whole studied group was 41.3 ±11.68 years and females accounted for over 2/3 of all subjects. Both obese and non-obese patients didn't fulfill the criteria for metabolic syndrome [31]. Although biochemical parameters had mean values within the normal range in the two subgroups, there were significant differences (p < 0.05) for plasma fasting glucose (88.32 ± 8.80 vs 99.28 ± 14.62 mg/dL, p = 0.0026), uric acid (5.29 ± 1.48 vs 7.9 ± 2.19 mg/dL, p = 0.0067) and hs-CRP (0.45 ± 0.056 vs 0.67 ± 0.019 mg/L, p = 0.0422). The most relevant aspect was that similar to other clinical studies in obesity [12-13, 37-38], plasma chemerin levels were significantly higher in obese patients compared to the control group (11.61 ± 2.31 vs 8.98 ± 1.89 ng/mL, p = 0.0004). It is worth mentioning that serum chemerin levels in our study were significantly lower compared to other reported series. Plasma adiponectin levels were also significantly different between the two subgroups (18.05 ± 1.55 vs 16.36 ± 1.49 ng/mL, p = 0.0003). Our results provided higher values for morbid obesity compared to similar studies [39]. This finding could be an argument for the direct correlation of serum adiponectin with the amount of visceral fat than subcutaneous fat and its biological functions [40]. For more in-depth study of adipokines contribution to subclinical ATS

we also studied the chemerin/adiponectin ratio. Chu et al. demonstrated the clinical significance of this ratio when the risk of metabolic syndrome was studied [24]. Our study demonstrated a significant higher value in obese subgroup (0.67 ± 0.18 vs 0.55 ± 0.12, p = 0.0052). Regarding the parameters of subclinical ATS, both subgroups had values within the normal range, except the high mean value of epicardial fat thickness (EFT) in obese subjects (5.3 ± 0.13 mm). We found significant differences between subgroups for SBPao (119.70 ± 20.18 vs 128.74 ± 20.81 mmHg, p = 0.011), mean IMTc (0.57 ± 0.07 vs 0.72 ± 0.21 mm, p = 0.033) and EFT (5.3 ± 0.13 vs 3.2 ± 0.08 mm, p= 0.0001).

The relation of plasma chemerin with subclinical ATS is controversial. Although the meta-analysis of Li et al. suggests a pathophysiological role of chemerin in obesity [17], clinical studies are conflicting [13-16, 18]. Data regarding the correlation of plasma chemerin with noninvasive markers of subclinical ATS in humans are limited and discordant [26-29]. Our data are synthetized for the whole sample and obese subjects in table 2.

Our results confirmed the positive correlation of chemerin to PWV in obese patients (r = 0.432, p = 0.035) and the whole sample (r = 0.272, p = 0.006) and are consistent with another clinical studies [26-28]. Regarding the other parameters of arterial stiffness we found that chemerin was related to SBPao (r = 0.459, p = 0.024), PPao (r = 0.414, p = 0.044) and SEVR (r = -0.409, p = 0.047), but not to Aix (p > 0.05). The negative correlation with SEVR (r = -0.409, p = 0.047), a predictor of coronary flow reserve, could suggest a potential role of chemerin on coronary microcirculation. Our study couldn't sustain the correlation with EFT in obese patients (p = 0.354) similar to a very recent study in prediabetes and diabetes [41]. On the other hand, these results are discordant with other clinical studies [as cited in Aydin et al. – 41] and also with our results in the whole sample (r = 0.369, p = 0.0001). This is possibly due to the study of different populations or of the local adipose tissue chemerin. Finally, we detected a positive correlation of chemerin with mean IMTc, stronger in obese subjects (r = 0.404, p = 0.05) compared to the whole sample (r = 0.28, p = 0.006). Our results were discordant with the study of Yoo et al. in apparently healthy obese patients [26], possible explained by the higher BMI in our study. Also, some clinical studies in non-complicated type 2 diabetes are consistent with our findings [42]. Using multiple linear regression models with chemerin as the dependent variable in obese patients, this adipokine was still not a predictor for mean IMTc, but remained an independent risk factor for PWV (r^2 = 0.88, p = 0.0001), SBPao (r^2 = 0.91, p = 0.0001), SEVR (r^2 = 0.53, p = 0.001) and also for aortic and brachial Aix 75 (r^2 = 0.85, p = 0.0001; r^2 = 0.71, p = 0.001, respectively). Regarding chemerin/adiponectin ratio we observed that it is not a superior parameter compared to chemerin itself to predict subclinical ATS. We have not a clear explanation for this finding. Most probably the levels of both adipokines and the characteristics of study population influenced the results. On the other hand, the reported results between studies are difficult to interpret, as long as clear cut-off values or reference ranges in specific populations are not available. The standardization of the measuring method of adipokines for research and clinical purposes could be also a key point.

To our knowledge this is the first study that provides information about the relation between circulating chemerin and all validated markers of arterial stiffness in apparently healthy obese subjects. However, the small size of sample and the method of chemerin measurement, not

Variable	Mean values			p-value
	Whole sample	Non-obese (control group)	Obese	
Age (years)	41.3 ±11.68	43.36 ± 13.9	39.24 ± 8.74	**0.021**
Female sex (%)	76	68	84	
BMI (kg/m²)	33.33 ± 10.88	24.24 ± 3.15	43.9 ± 6.07	**0.0001**
Waist circumference (cm)	104.27 ± 25.85	83.04 ± 8.75	125.5 ± 18.68	**0.0001**
WHR	0.89 ± 0.11	0.83 ± 0.08	0.96 ± 0.10	**0.0001**
Index central obesity	0.62 ± 0.14	0.50 ± 0.06	0.75 ± 0.08	**0.0001**
Systolic BP (mmHg)	123.7 ± 13.53	118.04 ± 11.72	129.36 ± 13.03	**0.0022**
Diastolic BP (mmHg)	71.18 ± 10.40	67.08 ± 7.89	75.28 ± 11.12	**0.0044**
Cholesterol (mg/dL)	199.60 ± 34.70	197.80 ± 41.39	201.4 ± 27.17	0.718
HDL-cholesterol (mg/dL)	50.18 ± 12.58	50.36 ± 14.94	50 ± 9.98	0.92
LDL-cholesteol (mg/dL)	126.36 ± 31.27	125.04 ± 39.97	127.68 ± 23.48	0.76
Triglycerides (mg/dL)	122.78 ± 21.64	121.24 ± 25.74	124.32 ± 17.96	0.86
Fasting plasma glucose (mg/dL)	93.80 ± 13.06	88.32 ± 8.80	99.28 ± 14.62	**0.0026**
Uric acid (mg/dL)	6.04 ± 2.0	5.29 ± 1.48	6.79 ± 2.19	**0.0067**
Creatinine (mg/dL)	0.76 ± 0.11	0.74 ± 0.09	0.78 ± 0.12	0.19
eGFR (ml/min/1.73 m²)	103.1 ± 13.87	105.22 ± 13.19	101.06 ± 14.09	0.29
hs-CRP (mg/L)	0.73 ± 0.096	0.45 ± 0.056	0.67 ± 0.019	**0.0422**
Chemerin (ng/mL)	10.27 ± 2.47	8.98 ± 1.89	11.61 ± 2.31	**0.0004**
Adiponectin (ng/mL)	17.20 ± 1.73	16.36 ± 1.49	18.05 ± 1.55	**0.0003**
Chemerin/adiponectin x 10⁻³	0.61 ± 0.16	0.55 ± 0.12	0.67 ± 0.18	**0.0052**
PWV (m/s)	9.26 ± 2.27	8.92 ± 2.14	9.59 ± 2.38	0.3
Aortic Aix (%)	29.6 ± 15.2	35.1 ± 16.2	24.1 ± 12.1	0.090
Aortic Aix 75 (%)	26.9 ± 13.6	30.3 ± 14.9	23.4 ± 11.4	0.072
Brachial Aix (%)	-16.1 ± 3.01	-5.5 ± 0.32	-26.7 ± 0.24	0.110
Brachial Aix 75 (%)	-18.90 ± 2.79	-10.41 ± 0.3	-27.40 ± 0.22	**0.03**
SBPao (mmHg)	124.08 ± 20.83	119.70 ± 20.18	128.74 ± 20.81	**0.011**
PPao (mmHg)	51.46 ± 11.67	50.66 ± 12.69	52.26 ± 10.76	0.63
SEVR	1.13 ± 0.24	1.19 ± 0.28	1.06 ± 0.16	0.054
Mean IMTc (mm)	0.65 ± 0.17	0.57 ± 0.07	0.72 ± 0.21	**0.0033**
Epicardial fat thickness (mm)	4.2 ± 0.15	3.2 ± 0.08	5.3 ± 0.13	**0.0001**

Table 1
BASELINE CHARACTERISTICS OF THE STUDY POPULATION

Subclinical ATS	Chemerin (ng/mL)		Chemerin/adiponectin ratio	
	Whole sample	Obese	Whole sample	Obese
PWV (m/s)	*r = 0.272 **p = 0.006	r = 0.432 p = 0.035	r = 0.218 p = 0.026	r = 0.324 p = 0.123
Aortic Aix (%)	r = -0.065 p = 0.512	r = 0.181 p = 0.398	r = -0.064 p = 0.509	r = 0.179 p = 0.403
Aortic Aix 75 (%)	r = 0.058 p = 0.558	r = 0.325 p = 0.121	r = 0.030 p = 0.757	r = 0.281 p = 0.184
Brachial Aix (%)	r = -0.058 p = 0.558	r = 0.181 p = 0.396	r = -0.064 p = 0.509	r = 0.180 p = 0.401
Brachial Aix 75 (%)	r = -0.01 p = 0.993	r = 0.261 p = 0.217	r = -0.020 p = 0.841	r = 0.238 p = 0.263
SBPao (mmHg)	r = 0.287 p = 0.004	r = 0.459 p = 0.024	r = 0.23 p = 0.02	r = 0.442 p = 0.031
PPao (mmHg)	r = 0.155 p = 0.119	r = 0.414 p = 0.044	r = 0.113 p = 0.248	r = 0.422 p = 0.040
SEVR	r = -0.259 p = 0.009	r = -0.409 p = 0.047	r = -0.214 p = 0.028	r = -0.381 p = 0.66
Mean IMTc (mm)	r = 0.28 p = 0.006	r = 0.404 p = 0.05	r = 0.239 p = 0.017	r = 0.447 p = 0.025
Epicardial fat thickness (mm)	r = 0.369 p = 0.0001	r = 0.198 p = 0.354	r = 0.276 p = 0.005	r = 0.030 p = 0.886

*r = correlation coefficient; **p = Sig. (2-tailed)

Table 2
PEARSON'S AND KENDALL TAU'S CORRELATION ANALYSIS OF CHEMERIN AND CHEMERIN/ADIPONECTIN LEVELS WITH THE PARAMETERS OF SUBCLINICAL ATS

specifically addressed to the different isoforms, are limitations of this research.

Conclusions

Chemerin could be considered a predictor of subclinical ATS. Defining its relation to the non-invasive biomarkers of subclinical ATS could be a more effective approach to early identification of obese patients at risk for cardio-metabolic disease. The conflicting results in literature suggest that chemerin has no direct vascular effect, but rather mediated by metabolic factors and mechanisms involved in early ATS such as inflammation or oxidative stress.

References

1.LIM, S., MEIGS, J.B. Links between ectopic fat and vascular disease in humans. Arterioscler. Thromb. Vasc. Biol. **34**, 2014, p 1820.

2.BROWN, N.K., ZHOU, Z., ZHANG, J., et al. Perivascular adipose tissue in vascular function and disease: a review of current research and animal models. Arterioscler. Thromb. Vasc. Biol. **34**, 2014, p 621.

3.YOO, H.J., CHOI, K.M. Adipokines as a novel link between obesity and atherosclerosis. World. J. Diab. **5**, no.3, 2014, p 357.

4.NTAIOS, G., GATSELIS, N.K., MAKARITSIS, K., DALEKOS, G.N. Adipokines as mediators of endothelial function and atherosclerosis. Atheroscler. **227**, 2013, p 216.

5.VILLACORTA, L., CHANG, L. The role of perivascular adipose tissue in vasoconstriction, arterial stiffness, and aneurysm. Horm. Mol. Biol. Clin. Investig. **21**, no.2, 2015, p 137.

6.WINDHAM, B.G., GRISWOLD, M.E., FARASAT, S.M., et al. Influence of leptin, adiponectin, and resistin on the association between abdominal adiposity and arterial stiffness. Am. J. Hypertens. **23**, 2010, p 501.

7.SHAH, A.S., DODAN, L.M., LAUER, A., et al. Adiponectin and arterial stiffness in youth type 1 diabetes patients: the SEARCH for diabetes in youth subjects. J. Pediatr. Endocrinol. Metab. **5**, 2012, p 717.

8.MATTERN, A., ZELLMANN, T., BECK-SICKINGER, A.G. Processing, Signaling, and Physiological Function of Chemerin. IUBMB. Life. **66**, no.1, 2014, p 19.

9.GORALSKI, K.B., MCCARTHY, T.C., HANNIMAN, E.A., et al. Chemerin, a novel adipokine that regulates adipogenesis and adipocyte metabolism. J. Biol. Chem. **282**, 2007, p 28175.

10.ROUGER, L., DENIS, R., LUANGSAY, S., PARMENTIER, M. ChemR23 knockout mice display mild obesity but no deficit in adipocyte differentiation. J. Endocrinol. **219**, 2013, p 279.

11.CORONA-MERAZ, F.I., NAVARRO-HERNANDEZ, R.E., RUIZ-QUEZADA, S.L., et al. Inverse Relationship of the CMKLR1 Relative Expression and Chemerin Serum Levels in Obesity with Dysmetabolic Phenotype and Insulin Resistance. Mediators of Inflammation. **2016**, ID 3085390, 2016, p 1.

12.SELL, H., DIVOUX, A., POITOU, C., et al. Chemerin correlates with markers for fatty liver in morbidly obese patients and strongly decreases after weight loss induced by bariatric surgery. J. Clin. Endocrinol. Metab. **95**, 2010, p :2892.

13.BOZAOGLU, K., BOLTON, K., MCMILAN, J., et al. Chemerin is a novel adipokine associated with obesity and metabolic syndrome. Endocrinology. **148**, 2007, p 4687.

14.KIM, S.H, LEE, S.H, AHN, K.Y, et al. Effect of lifestyle modification on serum chemerin concentration and its association with insulin sensitivity in overweight and obese adults with type 2 diabetes. Clin. Endocrinol. (Oxf). **80**, 2014, p 825.

15.CHAKAROUN, R., RASCHPICHLER, M., KLOTING, N., et al. Effects of weight loss and exercise on chemerin serum concentrations and adipose tissue expression in human obesity. Metabolism. **61**, 2012, p 706.

16.RESS, C., TSCHONER, A., ENGL, J., et al. Effect of bariatric surgery on circulating chemerin levels. Eur. J. Clin. Invest. **40**, 2010, p 277.

17.LI, Y., SHI, B., LI, S. Association between serum chemerin concentrations and clinical indices in obesity or metabolic syndrome: A Meta-Analysis. PLoS. One. **9**, no.12, 2014, p 1.

18.BECKER, M., RABE, K., LEBHERZ, C., et al. Expression of human chemerin induces insulin resistance in the skeletal muscle but does not affect weight, lipid levels, and atherosclerosis in LDL receptor knockout mice on high-fat diet. Diabetes. **59**, 2010, p 2898.

19.LEHRKE, M., BECKER, A., GREIF, M., et al. Chemerin is associated with markers of inflammation and components of the metabolic syndrome but does not predict coronary atherosclerosis. Eur. J. Endocrinol. **161**, 2009, p 339.

20.NEVES, K.B., DINH, CAT, A.N., MOREIRA, LOPES, R.A., et al. Chemerin Regulates Crosstalk Between Adipocytes and Vascular Cells Through Nox. Hypertension. **66**, no.3, 2015, p 657.

21.RHEE, E.J. Chemerin: a novel link between inflammation and atherosclerosis? Diabetes. Metab. **35**, 2011, p 216.

22.KOSTOPOULOS, C.G., SPIROGLOU, S.G., VARAKIS, J.N., et al. Chemerin and CMKLR1 expression in human arteries and periadventitial fat: a possible role for local chemerin in atherosclerosis? BMC. Cardiovasc. Dis. **14**, no.1, 2014, p 56.

23.SINAN, Y., GÖKHAN, A., PINAR, D. Chemerin as an independent predictor of cardiovascular event risk. Ther. Adv. Endocrinol. Metab. **7**, no.2, 2016, p 57.

24.CHU, S.H., LEE, M.K., AHN, K.Y., et al. Chemerin and Adiponectin Contribute Reciprocally to Metabolic Syndrome. PLoS. One. **7**, no.4, 2012, p 1.

25.COIMBRA, S., BRANDAO PROENCA, J., SANTOS-SILVA, A., NEUPARTH, M.J. Adiponectin, leptin, and chemerin in elderly patients with type 2 diabetes mellitus: a close linkage with obesity and length of the disease. Biomed. Res. Int. **2014**, no 701915, 2014, p 1.

26.YOO, H.J., CHOI, H.Y., YANG, S.J., et al. Circulating chemerin level is independently correlated with arterial stiffness. J. Atheroscler. Thromb. **19**, 2012, p 59.

27.ALI, T.M., HADIDI, K.A. Chemerin is associated with markers of inflammation and predictors of atherosclerosis in Saudi subjects with metabolic syndrome and type 2 diabetes mellitus. Beni-Suef Univ. J. Basic. Appl. Sci. **2**, 2013, p 86.

28.GU, P., CHENG, M., HUI, X., et al. Elevating circulation chemerin level is associated with endothelial dysfunction and early atherosclerotic changes in essential hypertensive patients. J. Hypertens. **33**, no.8. 2015, p 1624.

29.AYDIN, K., CANPOLAT, U., SAFAK, A., et al. Chemerin is not associated with subclinical atherosclerosis markers in prediabetes and diabetes. Anatol. J. Cardiol. **16**, 2016, p 749.

30.GAO, X., MI, S., ZHANG, F., et al. Association of chemerin mRNA expression in human epicardial adipose tissue with coronary atherosclerosis. Cardiovascular. Diabetology. **10**, 2011, p 87.

31.ALBERTI, K.G.M.M., ECKEL R.H., GRUNDY, S.M., et al. Harmonizing the Metabolic Syndrome. Circulation. **120**, 2009, p 1640.

32.ZABEL, B.A., KWITNIEWSKI, M., BANAS, M., et al. Chemerin regulation and role in host defense. Am. J. Clin. Exp. Immunol. **3**, no.1, 2014, p 1.

33.NIGRO, E., SCUDIERO, O., MONACO, M.L., et al. New Insight into Adiponectin Role in Obesity and Obesity-Related Diseases. BioMed. Research. International. **2014**, 2014, p 1.

34.VAN BORTEL, L.M., LAURENT, S., BOUTOUYRIE, P., et al. Expert consensus document on the measurement of aortic stiffness in daily practice using carotid-femoral pulse wave velocity. J. Hum. Hypertens. **26**, no1, 2012, p 64.

35.TOUBOUL, P.J., VICAUT, E., LABREUCHE, J., et al. Mannheim carotid intima-media thickness and plaque consensus. Cerebrovasc. Dis. **34**, no.4, 2012, p 290.

36.BERTASO, A.G., DANIELA BERTOL, D., DUNCAN, B.B., FOPPA, M. Epicardial Fat: Definition, Measurements and Systematic Review of Main Outcomes. Arq. Bras. Cardiol. **101**, no. 1, 2013, p 18.

37.CHANG, S.S., EISENBERG, D., ZHAO, L., et al. Chemerin activation in human obesity. Obesity (Silver Spring). **24**, no.7, 2016, p 1522

38.BOZAOGLU, K., SEGAL, D., SHIELDS, K.A., et al. Chemerin is associated with metabolic syndrome phenotypes in a Mexican-American population. J. Clin. Endocrinol. Metab. **94**, 2009, p 3085.

39.GELONEZE, B., PEREIRA, J.A., PAREJA, J.C., et al. Overcoming metabolic syndrome in severe obesity: adiponectin as a marker of insulin sensitivity and HDL-cholesterol improvements after gastric bypass. Arq. Bras. Endocrinol. Metab. **53**, no.2, 2009, p 293.

40.KISHIDA, K., KIM, K.K., FUNAHASHI, T., et al. Relationships between circulating adiponectin levels and fat distribution in obese subjects. J. Atheroscler. Thromb. **18**, no.7, 2011, p 592.

41.AYDIN, K., CANPOLAT, U., AKIN, S., DURAL, M. Chemerin is not associated with subclinical atherosclerosis markers in prediabetes and diabetes. Anatol. J. Cardiol. **16**, 2016, p 749.

42.KORANI, M.A., BADR, E.A.E., ZEITON, A.A. Serum chemerin level and its relation to carotid intima media thickness in type 2 diabetic patients. J. Adv. Biol. **7**, no.3, 2015, p 1414.