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Factors associated with atherosclerotic plaque echogenicity in patients aged 40-64 with carotid atherosclerosis

Aim To identify clinical and laboratory indexes related with the atherosclerotic plaque (ASP) echogenicity based on results of the analysis of grey-scale median (GSM) in patients aged 40–64 years.

Material and methods The study included patients aged 40–64 years with carotid atherosclerosis. The carotid duplex scanning was performed for all patients. The GSM analysis of obtained images was performed with the Adobe Photoshop CS6 software.

Results Atherosclerotic cardiovascular diseases were found in 31 (21.4%) patients. Correlation analysis determined inverse interrelationships between GSM and the body weight index (BWI) (r=–0.359; p<0.0001), waist circumference (r=–0.357; p<0.0001), and levels of uric acid (r=–0.244; p=0.021) and glucose (r=–0.205; p=0.032). According to the regression, statistically significant correlations remained between GSM and BWI as well as the waist circumference after the adjustment for sex and age.

Conclusion In patients with carotid atherosclerosis aged 40–64 years, the decrease in ASP GSM was associated with increases in BWI and waist circumference.

Keywords Atherosclerotic plaque; atherosclerotic plaque echogenicity; GSM analysis

For citation Genkel V.V., Kuznetsova A.S., Lebedev E.V., Shaposhnik I.I. Factors associated with atherosclerotic plaque echogenicity in patients aged 40-64 with carotid atherosclerosis. Kardiologiia. 2021;61(6):35–40. [Russian: Генкель В.В., Кузнецова А.С., Лебедев Е.В., Шапошник И.И. Факторы, связанные с эхогенностью атеросклеротических бляшек, у пациентов в возрасте 40–64 лет с каротидным атеросклерозом. Кардиология. 2021;61(6):35–40]

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Introduction

According to large-scale epidemiological studies conducted among other countries within the Russian Federation, subclinical carotid atherosclerosis defined by the presence of an atherosclerotic plaque is a predictor of adverse cardiovascular events [1]. An active search is underway to determine new ultrasound markers of carotid atherosclerosis which will provide additional diagnostic and prognostic information after detecting plaques and determining the degree of carotid stenosis [2].

Quantitative analysis of the plaque echogenicity by determining the grayscale median (GSM) allows non-invasive evaluation of the atheroma morphology to be performed and unstable plaques to be identified during duplex scanning of vessels [3]. Hypoechoic plaques (GSM <30) with an extensive lipid necrotic core are predictors of adverse cardiovascular events [4–7]. Identification of factors associated with plaque echogenicity is important both in terms of the optimization of diagnosis algorithms and monitoring of patients with subclinical atherosclerosis, as well as with regard to understanding the mechanisms of plaque destabilization. There is limited data available on this issue. According to the GSM analysis carried out under the Multi-Ethnic Study of Atherosclerosis (MESA), male sex, age, and ethnicity (GSM was lower in African American patients) were associated with the echogenicity of carotid plaques [8]. GSM was not statistically significantly correlated with other factors. The Northern Manhattan Study (NOMAS) demonstrated that smoking more than doubles the probability of detecting a plaque with GSM<48 (the first quintile) [9].

Objective

To identify clinical and laboratory parameters associated with the plaque's echogenicity according to the GSM analysis in patients aged 40 to 64.

Material and methods

The study included patients aged 40 to 64 with carotid atherosclerosis defined by the presence of plaques in the carotid arteries. The source population consisted of patients referred for carotid duplex scanning to assess cardiovascular risk during outpatient care. Signed informed consent was the inclusion criterion. The study was performed following the World Medical Association's Declaration of Helsinki on ethical principles for medical research involving human subjects developed originally in 1964 and amended in 2000. The study protocol was approved by the ethics committee of the South Ural State Medical University (Minutes No. 10

dated 27/10/2018). The exclusion criteria were severe liver and kidney failure (glomerular filtration rate (GFR) less than 30 mL/min/1.73 m^2) and malignancies.

Ultrasound examination

All patients underwent carotid duplex scanning. The following vessels were inspected bilaterally in the longitudinal and cross-section views: common carotid arteries (CCAs) and CCA bifurcation; internal carotid arteries (ICAs); and external carotid arteries (ECAs) from the anterior, lateral, and posterior views. The examination was conducted using a 10 MHz linear probe frequency on a Samsung Medison EKO7 digital multifunction scanner (Republic of Korea). Focal thickening of the intima media of more than 1.5 mm or by 0.5 mm more than the surrounding intima-media thickness (IMT), or by 50% more than IMT of the adjacent areas of CCAs was defined as a plaque [10]. The percent of stenosis was measured in the planimetric B-mode by a cross-sectional diameter of the vessel following the method developed in the European Carotid Surgery Trial (ECST). Maximum stenosis was measured in a particular patient [11].

The longitudinal view allowed for the best imaging of atheroma, and the image was then saved and exported. GSM analysis of the images was performed using Adobe Photoshop CS6 (Adobe System Incorporated, USA) following the standard procedure [4, 12, 13]. The imported image was normalized, taking the vessel lumen as the black reference values and adventitia as the white reference values [14]. The modified GSM was then determined in the area of interest corresponding to the plaque. The stages of the GSM analysis are shown in Figure 1.

Body mass index (BMI) was calculated by the Quetelet formula: weight (kg)/height (m^2). BMI $\geq$ 30 kg/m^2 was considered obese. Waist circumference (RT) was measured using the standard method with a measuring tape in the upright position, arms down to the sides. Abdominal obesity was defined as >94 cm for male patients and >80 cm for female patients.

Laboratory investigations

The following biochemical blood parameters were determined after at least 8 hours of fasting: total cholesterol (TC); low-density lipoprotein cholesterol (LDL–C); high-density lipoprotein cholesterol (HDL–C); triglycerides (TG); glucose; glycolated hemoglobin; uric acid; and creatinine with the subsequent calculation of glomerular filtration rate (GFR) using the CKD-EPI formula.

Statistical analysis

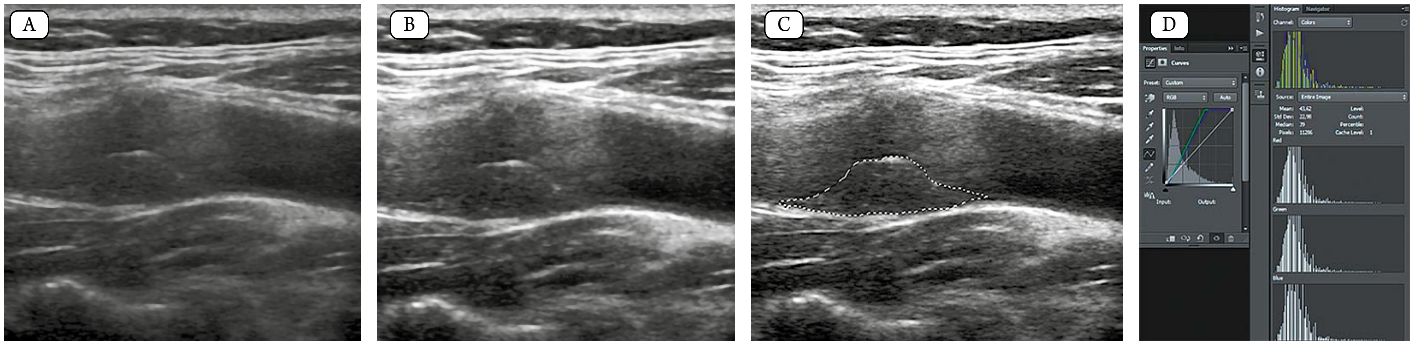
The statistical analysis of the data obtained was performed using SPSS Statistics version 18. The qualitative variables were described using the absolute and relative rates (percentages). The quantitative variables were expressed using the median (Me) and the interquartile range [25th percentile; 75th percentile] if the distribution was non-normal. If the distribution was normal, the mean (M) and standard deviation (SD) were used. The correlations of indicators were determined using the Spearman correlation coefficient. The significance of differences between the two groups was assessed using the Mann-Whitney test. The differences were considered statistically significant with p=0.05. Linear regression was used to estimate the dependence of one quantitative variable on another.

Results

The study included 145 patients with carotid atherosclerosis: 79 (54.5%) male patients and 66 (45.5%) female patients. Clinical and laboratory characteristics of patients are provided in Table 1.

Thus, atherosclerotic cardiovascular diseases were established in 31 (21.4%) patients. It should also be noted that drug therapy is indicated as at the time of inclusion, i.e.

Figure 1. GSM-analysis of plaques



A — exported native image; B — the normalized image of a plaque;
C — mapping the area of interest corresponding to the plaque contours; D — determining the grayscale median.

ISSN 0022-9040. Kardiologiia. 2021;61(6). DOI: 10.18087/cardio.2021.6.n1536

Table 1. Clinical and laboratory characteristics of patients

Parameters	Patients, n = 145
Age, years, Me (IR)	53.0 (46.0; 59.0)
BMI, kg/m2, Me (IR)	27.1 (24.8; 30.5)
Obesity, n (%)	43 (29.6)
Waist circumference, cm, Me (IR)	90.0 (83.0; 98.0)
Abdominal obesity, n (%)	85 (58.6)
Smoking, n (%)	23 (15.8)
Arterial hypertension, n (%)	89 (61.4)
Coronary artery disease, n (%)	27 (18.6)
Myocardial revascularization, n (%)	11 (7.58)
Postinfarction cardiosclerosis, n (%)	13 (8.96)
CVA, n (%)	2 (1.37)
Intermittent claudication, n (%)	2 (1.37)
Antiplatelet agents, n (%)	33 (22.7)
Beta blockers, n (%)	42 (28.9)
RAAS inhibitors, n (%)	51 (35.2)
Diuretics, n (%)	19 (13.1)
Statins, n (%)	51 (35.1)
TC, mmol/L, Me (IR)	6.19 (5.12; 7.10)
LDL-C, mmol/L, Me (IR)	3.96 (3.01; 4.73)
HDL-C, mmol/L, Me (IR)	1.32 (1.15; 1.61)
TG, mmol/L, Me (IR)	1.37 (1.00; 1.90)
Glucose, mmol/L, Me (IR)	5.60 (5.11; 6.13)
Glycolated hemoglobin, %, Me (IR)	5.33 (5.20; 5.97)
GFR, mL/min/1.73m^2, Me (IR)	71.0 (62.0; 89.5)

BMI, body mass index; DM, diabetes mellitus; RAAS, renin-angiotensin-aldosterone system; TC, total cholesterol; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglycerides; GFR, glomerular filtration rate, CVA, cerebrovascular accident; IR, interquartile range.

before the carotid duplex scanning results were obtained (Table 2).

Figure 2 shows the distribution of patients depending on the plaque GSM values. Thus, hypoechoic plaques (GSM<30) were detected in 13 (8.96%) patients. The GSM values in patients receiving statins did not differ significantly from those of patients not receiving statins (p=0.282). Based on the correlation analysis, inverse correlations were established between GSM and BMI (r=–0.359; p<0.0001), WC (r=–0.357; p<0.0001), uric acid (r=–0.244; p=0.021), and glucose (r=–0.205; p=0.032). Linear regression analysis adjusted for age and gender was carried out, in order to

Table 2. Results of carotid duplex scanning

Parameters	Patients, n = 145
Carotid plaques, n (%)	145 (100)
LCCA stenosis, %, Me (IR)	23.5 (0.00; 30.5)
RCCA stenosis, %, Me (IR)	25.0 (0.00; 30.0)
LICA stenosis, %, Me (IR)	0.00 (0.00; 30.0)
RICA stenosis, %, Me (IR)	0.00 (0.00; 26.5)
Maximum stenosis, Me (IR)	30.0 (25.0; 38.5)
Carotid stenoses ≥50%, n (%)	16 (11.0)
GSM, Me (IR)	64.0 (47.0; 81.0)

CA, carotid artery; LCCA, left common carotid artery; RCCA, right common carotid artery; LICA, left internal carotid artery; RICA, right internal carotid artery; GSM, grayscale median, IR, interquartile range.

Figure 2. Distribution of patients according to the plaque GSM values

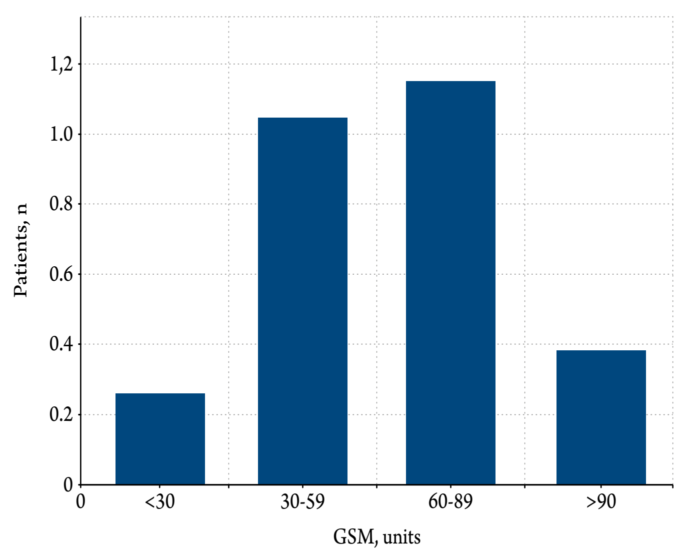


Table 3. Results of the gender and age adjusted linear regression analysis showing the correlations between GSM, BMI, and WC

Parameter	R	R2	B	95% CI for B		p
				Lower limit	Upper limit	
GSM						
BMI	0.374	0.140	-1.94	-2.85	-1.04	<0.0001
GSM						
WC	0.354	0.125	-0.65	-0.98	-0.31	<0.0001

CI, confidence interval; BMI, body mass index; WC, waist circumference.

Figure 3. Correlations between plaque GSM and body mass index

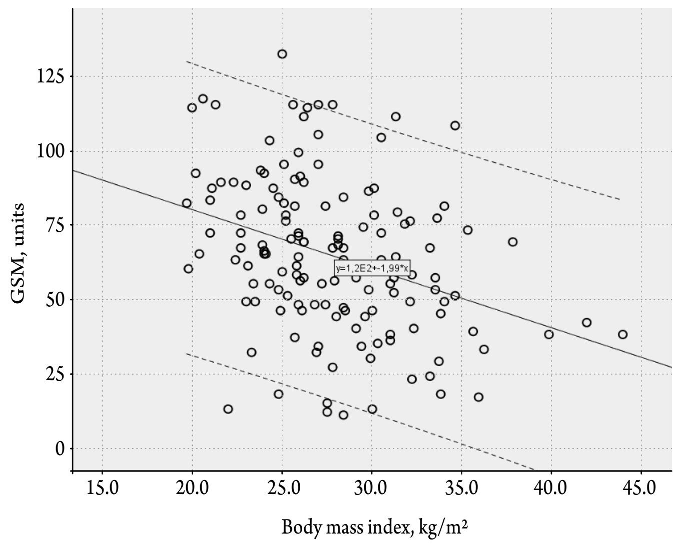
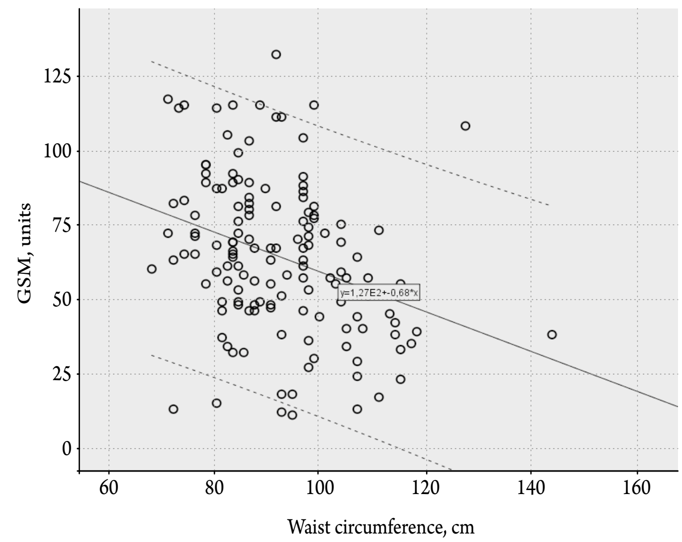


Figure 4. Correlations between plaque GSM and waist circumference



identify the independent correlations between GSM and these factors.The results are shown in Table 3.

According to the linear regression, a statistically significant correlation remained after adjustment for age and gender between GSM and BMI (Figure 3) and WC (Figure 4). Thus, BMI and WC were responsible for up to 14.0% of plaque GSM variability.

Discussion

The presence of hypoechoic carotid plaques is an independent predictor of adverse cardiovascular events, including stroke. Moreover, progressively decreasing echogenicity of plaques during watchful waiting provides additional prognostic information and allows patients at the highest risk of cardiovascular accidents to be identified [15].

The main result of the study is the establishment of independent correlations between the carotid plaque echogenicity on the one hand, and BMI and WC on the other. Moreover, correlations between GSM and the levels of uric acid and glucose were identified which were not significant in the age and gender adjusted linear regression analysis. Earlier, large clinical trials have demonstrated that obesity and a metabolic risk factor cluster are the most potent predictors of the presence of unstable and complicated plaques. Rovella et al. demonstrated that obesity was associated with a 5.91-fold increase (95% CI: 1.17–29.8) in the relative risk of unstable carotid plaques according to histomorphological analyses [16]. Earlier, Kadoglou et al. in their study of 74 patients with carotid atherosclerosis showed independent inverse correlations between body fat and carotid plaque echogenicity according to the GSM analysis [17]. Moreover, several studies have established the effects of obesity on the composition of plaque in different vascular systems according to the use of various imaging techniques [18]. The most significant mechanisms of correlations between obesity and plaque structures included the activation of inflammatory signaling pathways [19, 20].

It should thus be noted that obesity is a factor with a significant effect on the quantitative and qualitative characteristics of atherosclerotic lesions of vessels. For example, obesity is known to be an independent predictor of the progression of subclinical atherosclerosis, including

ISSN 0022-9040. Kardiologiia. 2021;61(6). DOI: 10.18087/cardio.2021.6.n1536

during statin therapy [21, 22]. Quantitative assessment of atherosclerosis burden and plaque echogenicity in obese patients may contribute to obtaining additional prognostic information in these patients. Segmental GSM analysis of carotid atheromas is also a promising method. It has already been demonstrated that the presence of hypoechoic foci in the plaque parts adjacent to the vessel lumen may have additional prognostic value [23, 24].

There is a sufficient number of publications which demonstrate correlation between the administration of statins and GSM [25]. However, we have not found significant differences in the GSM values depending on the use of statins. This result may be due to a number of reasons: failure to achieve the target levels of LDL–C: insufficient duration of statin therapy; and heterogeneity of the study population.

The possible limitations of this study are its small sample size, heterogeneity of the study population, and the single-center nature of the study.

Conclusion

In patientsaged 40 to 64 with carotid atherosclerosis, decreased plaque GSM values were associated with increased BMI, WC, uric acid, and glucose levels. According to the linear regression analysis, a statistically significant correlation remained after the adjustment for age and gender between GSM and BMI and WC.

No conflict of interest is reported.

The article was received on 20/01/2021

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ISSN 0022-9040. Kardiologiia. 2021;61(6). DOI: 10.18087/cardio.2021.6.n1536