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Disease Activity, Oxidized-LDL Fraction and Anti-Oxidized LDL Antibodies Influence Cardiovascular Risk in Rheumatoid Arthritis*

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A – research concept and design; **B** – collection and/or assembly of data; **C** – data analysis and interpretation; **D** – writing the article; **E** – critical revision of the article; **F** – final approval of article

Abstract

Background. Patients with rheumatoid arthritis (RA) have a shortened lifespan compared to the general population. The high rate of premature mortality in the RA population can be attributed to cardiovascular disease (CVD).
Objectives. The aim of the study was to look for non-classic risk factors that can at least partially explain the enhanced cardiovascular (CV) risk in patients with RA.
Material and Methods. This was an observational study with 37 RA patients and 24 healthy volunteers as controls. The participants' medical history was taken, and systematic coronary risk evaluation (SCORE) and carotid ultrasonography examinations were performed on all the participants. Laboratory tests included antibodies anti-cyclic citrullinated peptide (anti-CCP), inflammatory markers, lipid level, oxidized low-density lipoprotein (oxLDL) level and the level of anti-oxLDL antibodies.
Results. Both SCORE and oxLDL fraction were elevated in RA patients as compared to the healthy controls (3.1 ± 3.7 vs. 0.8 ± 1.2, p = 0.005; and 0.029 ± 0.033% vs. 0.014 ± 0.006%, p = 0.04, respectively). In the RA group, the presence of anti-CCP was associated with thickening of the carotid intima-media complex and SCORE elevation. In the RA group, significant correlations were found between SCORE and mean carotid intima-media thickness (IMT; RP = 0.34, p = 0.040), disease activity score (RP = 0.42, p = 0.011), erythrocyte sedimentation rate (ESR; RP = 0.35, p = 0.036), and disease duration (RP = 0.52, p = 0.002). In RA patients with carotid plaques, the oxLDL fraction was significantly elevated in comparison to those without plaques (0.055 ± 0.070% vs. 0.022 ± 0.018%, p = 0.033). In the RA group, there was a significant negative correlation between mean carotid IMT and the serum concentration of anti-oxLDL antibodies (RP = –0.38, p = 0.02). No association was noted between the presence of rheumatoid nodules and SCORE or carotid IMT.
Conclusions. Among RA patients, disease activity, ESR, disease duration, the presence of anti-CCP antibodies, the oxLDL fraction and the level of anti-oxLDL antibodies influence CV risk (**Adv Clin Exp Med 2016, 25, 1, 43–50**).

Key words: intima-media thickness, oxidized low-density lipoprotein cholesterol, anti-oxidized low-density lipoprotein cholesterol antibodies, rheumatoid arthritis, systematic coronary risk evaluation.

It is well documented that patients with rheumatoid arthritis (RA) have shortened lifespans compared to the general population [1] and that the mortality gap between RA patients and the general population is continuously widening [2]. Many publication have demonstrated that the high premature mortality in the RA population can be at least partially attributed to cardiovascular disease

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(CVD) [3–5]. The risk of CVD in RA patients is comparable to the risk in patients with type 2 diabetes mellitus [6]. However, the increased risk of CVD in RA population cannot be fully explained by traditional cardiac risk factors [7]. It has been reported recently that the CVD risk in women with RA is twofold higher than their Framingham score predicts, and in men with RA the risk is 65% higher than predicted [8]. Some authors report as much as a fourfold increase in cardiovascular (CV) events among RA patients [7]. Some of the non-traditional CVD risk factors mentioned are the inflammatory process, the presence of autoantibodies (rheumatoid factor [RF] and anti-citrullinated peptide [anti-CCP] antibodies) and corticosteroid usage are mentioned [7, 9, 10].

It has been suggested that inflammation that occurs in RA patients predisposes them to accelerated premature atherosclerosis [11]. The association between carotid atherosclerosis and inflammatory markers (the erythrocyte sedimentation rate [ESR] and C-reactive protein [CRP] levels) has been demonstrated to be independent of classical CVD risk factors [12]. The present study analyzes the association between atherosclerosis and Disease Activity Score 28 (DAS28).

In RA rheumatoid nodules are considered to be associated with severe aggressive disease. The present study therefore also investigates the association between atherosclerosis and rheumatoid nodules.

The formation of atherosclerotic plaque begins with the deposition of lipids in subendothelial space. All apolipoprotein B-containing lipoproteins, e.g. low-density lipoprotein (LDL), can enter subendothelial space and can convert macrophages that have taken them up into foam cells. However, LDL has to be oxidized (oxLDL) in order to induce foam cell formation [13]. Inflammatory cells involved in RA pathogenesis, such as macrophages and lymphocytes, stimulate reactive oxygen species, leading to increases in oxLDL levels. OxLDL induces the production of autoantibodies by B-cells, and anti-oxLDL antibodies are present in patients with atherosclerosis and in healthy individuals [14]. On the one hand, the association between anti-oxLDL antibodies and CVD has been demonstrated [15, 16]; on the other hand, some experimental data suggest that these antibodies may play a protective role [17].

Atherosclerosis is a slowly progressing inflammatory disease leading to severe CV complications. However, it remains asymptomatic for many years before the first CV event occurs. One of the first stages of subclinical atherosclerosis is thickening of the intima-media complex [18]. It has been demonstrated that thickening of the carotid intima-media complex measured by B-mode ultrasound is associated with future CV events [19]. Carotid intima-media thickness (IMT) and carotid plaques can be regarded as CV risk factors in non-rheumatic individuals as well as in RA patients [20]. The European League against Rheumatism (EULAR) recommends using a modified Systematic Coronary Risk Evaluation (SCORE) to determine the 10-year risk of fatal CVD in RA patients [5].

The aim of this study was to look for non-classical risk factors that can at least partially explain the increased CVD risk in RA patients. The risk of CV events was assessed on the basis of IMT measurements and a modified SCORE. Disease activity, the presence of rheumatic nodules and autoantibodies, the oxidation of LDL and the production of anti-oxLDL antibodies were analyzed.

Material and Methods

Participants

The study was conducted on 37 patients (34 females, 3 males) aged 20–72 years (mean age 50.1 years) with an established diagnosis of rheumatoid arthritis. The inclusion criteria for the RA group were an established diagnosis of RA and age above 18 years. Exclusion criteria for the RA group included overlap syndromes, peripheral arterial disease (PAD), coronary heart disease (CHD), heart failure, cardiomyopathy, a history of cerebral stroke, severe renal dysfunction, chronic liver failure, chronic or acute infections and a history of malignant neoplasm.

As a control group 24 healthy volunteers (21 females, 3 males) aged 34–62 years (mean age 48.1 years) were enrolled. The exclusion criteria for the control group included RA or any other arthritis or connective tissue disease, cardiovascular diseases, any other organ system disease, chronic or acute infections and a history of malignant neoplasm. The volunteers were also excluded if physical examination revealed any clinically significant abnormalities.

The study was approved by the Wroclaw Medical University ethics committee (05.05.2010; No. KB-153/2011) and written informed consent was obtained from all participants prior to their inclusion in the study.

Clinical Data

The data for the analysis were obtained from medical histories, physical examinations, laboratory tests and ultrasound examinations of the carotid

arteries. The participants' data were collected, including age, sex, body mass index (BMI), systolic blood pressure (SBP), diastolic blood pressure (DBP), smoking habits, medical history, and current medication. Hypertension was defined as an SBP greater than 140 mm Hg and/or DBP greater than 90 mm Hg on repeated measurements, and/or receiving antihypertensive treatment. Hyperlipidemia was defined as total cholesterol (TC) concentration greater than 5.2 mmol/L and/or low-density lipoprotein cholesterol (LDL-C) concentration greater than 3.5 mmol/L, and/or high-density lipoprotein cholesterol (HDL-C) concentration lower than 0.9 mmol/L for males and 1.3 mmol/L for females, and/or triglycerides (TG) concentration greater than 2.3 mmol/L, and/or being under hypolipidemic treatment.

In the RA group, disease activity was measured using the Disease Activity Score 28 (DAS28). High disease activity was defined as a DAS28 score ≥ 5.1, low disease activity was defined as DAS28 < 3.2, and remission as DAS28 < 2.6.

SCORE Risk Estimation

The SCORE for the control group and the EULAR-modified SCORE for the RA group were calculated to determine the 10-year risk of fatal CV disease [5, 21]. The SCORE assessment was based on gender, age, smoking, systolic blood pressure and atherogenic index (total cholesterol/high-density lipoprotein cholesterol) [5, 21]. In accordance with EULAR's recommendation, a multiplication factor of 1.5 was used when a patient with RA met two of the following three criteria: disease duration of more than 10 years, the presence of RF or anti-CCP antibodies, and/or the presence of extra-articular manifestations [5].

Carotid Ultrasonography Examination

The IMT in the common carotid artery (CCA) was measured according to accepted methodology [22], using a GE Vivid 7 Dimension ultrasound device (General Electric) equipped with a 12-MHz linear-array transducer and an automatic protocol for IMT measurement applying gray-scale analysis. During the ultrasonography examination focal plaques in the extracranial carotid tree were also detected [22]. Plaques were defined as IMT ≥ 1.3 mm.

Salonen et al. observed that a mean carotid IMT > 0.70 mm was associated with increased risk of myocardial infarction (MI), and that the risk of MI increased 11% per 0.10 mm of IMT [23]. In the present study thickening of intima media was therefore defined as carotid IMT > 0.70 mm.

IMT > 0.90 mm and the presence of plaques are predictors of CV events in RA patients and in the general population [20, 24]. In the present study high/very high CV risk was therefore defined as IMT > 0.90 mm and/or the presence of plaques.

Laboratory Measurements

A venous blood sample was collected from each participant under fasting conditions. TC, HDL-C, LDL-C and TG concentrations in both groups were measured in a certified commercial laboratory. In the RA group, the CRP level and ESR were measured as well. In the RA group, the presence of RF and anti-CCP antibodies was recorded. The serum concentration of oxLDL and anti-oxLDL antibodies was measured by an enzyme-linked immunosorbent assay (ELISA) using commercially available kits (ox-LDL/MDA AdductELISA Kit and Anti ox-LDL ELISA Kit, Immundiagnostik AG, Bensheim, Germany). All the tests for each sample were performed according to the manufacturer's instructions in random order by a technician who was unaware of which group the sample belonged to. The oxidized LDL fraction in LDL was calculated.

Statistical Analysis

All numeric variables were expressed as mean ± standard deviation (SD), and categorical data were expressed as a number (n) and percentage in parentheses. Data were tested for normal distribution using the Kormogorov-Smirnov test. A univariate analysis of normally distributed continuous numerical variables was done with Student's *t* test, while the $\chi 2$ test was used for categorical variables. Correlations were assessed by the Pearson correlation analysis. All tests of significance were two-tailed. A p-value of less than 0.05 was considered statistically significant. All statistical analyses were performed with STATISTICA software, v. 10 (StatSoft, Inc, Tulsa, OK, USA).

Results

The characteristics of the study population are shown in Table 1. There was no difference between the groups in terms of age or the proportion of women. In the RA group, hypertension was observed in 38% of the patients, while in the control group no one was diagnosed as hypertensive. The rate of cigarette smoking was equivalent in the two groups, and RA patients were significantly

Table 1. Clinical characteristics of the participants

	Controls (n = 24)	**RA patients (n = 37)**	***P*[a]**
Females*	21 (87.5)	34 (91.9)	ns.
Age, years#	48.1 ± 7.6	50.1 ± 15.1	ns.
Concomitant diseases* • hypertension • hyperlipidemia	 0 18 (75.0)	 14 (37.8) 21 (56.8)	 0.0003 ns.
Smokers*	6 (25.0)	9 (24.3)	ns.
SCORE[b] #	0.8 ± 1.2	3.1 ± 3.7	0.005
RA duration, years#	NA	12.8 ± 10.3	
Autoantibodies* • RF • anti-CCP	 NA NA	 25 (67.6) 20 (54.1)	
Rheumatoid nodules *	NA	11 (29.7)	
DAS28#	NA	4.3 ± 1.45	
RA treatment • DMARDs* • Biologics* • GCs * • mean daily dose of GCs[c], mg#	 NA NA NA NA	 32 (86.5) 11 (29.7) 28 (75.7) 7.3 ± 4.5	
Mean IMT, mm#	0.57 ± 0.12	0.54 ± 0.18	ns.
IMT > 0.90 mm and/or carotid plaques* • carotid plaques*	6 (25.0) 2 (8.3)	8 (21.6) 6 (16.2)	ns. ns.
TC, mmol/L#	6.15 ± 1.10	5.65 ± 1.26	ns.
LDL-C, mmol/L#	3.91 ± 0.95	3.31 ± 1.04	0.03
HDL-C, mmol/L #	1.69 ± 0.33	1.66 ± 0.50	ns.
TG, mmol/L #	1.20 ± 0.49	1.53 ± 0.66	0.04
oxLDL fraction of LDL-C, % #	0.014 ± 0.006	0.029 ± 0.033	0.04
Anti-oxLDL antibodies,U/mL #	50700 ± 10500	52100 ± 21200	ns.

* Data is presented as number (percentage); # data is presented as mean ± standard deviation;
[a] RA group vs. control group; [b] EULAR-modified SCORE in RA group [7]; [c] counted as a dose of prednisone.

less likely to have elevated levels of LDL cholesterol. Although the mean carotid IMT was equivalent in the two groups, SCORE results were significantly higher in the RA group. In the RA group, significant elevation of the oxLDL fraction was also found.

In both the RA group and in the controls, significant correlations between the mean carotid IMT and age were detected (RP = 0.65, $p < 0.0001$ and RP = 0.54, $p = 0.008$, respectively).

Among the RA anti-CCP-positive patients, the carotid intima-media complex was significantly thicker than among the anti-CCP-negative ones (0.58 ± 0.18 mm vs. 0.38 ± 0.23 mm, $p = 0.039$) and the SCORE was elevated (4.7 ± 3.9 vs. 0.9 ± 1.3, $p = 0.049$). There was no association between IMT and ESR, CRP or the presence of RF.

In the RA group, significant correlations were found between SCORE results and the mean carotid IMT (RP = 0.34, $p = 0.040$), DAS28 (RP = 0.42, $p = 0.011$), ESR (RP = 0.35, $p = 0.036$), and disease duration (RP = 0.52, $p = 0.002$). Among the RA patients that were in remission or had low disease activity (11 patients), the SCORE was significantly lower than in RA patients with moderate to high disease activity (24 patients); (0.8 ± 1.8 vs. 4.0 ± 4.0, $p = 0.016$).

The mean carotid IMT and SCORE were equivalent in RA patients with and without rheumatoid nodules.

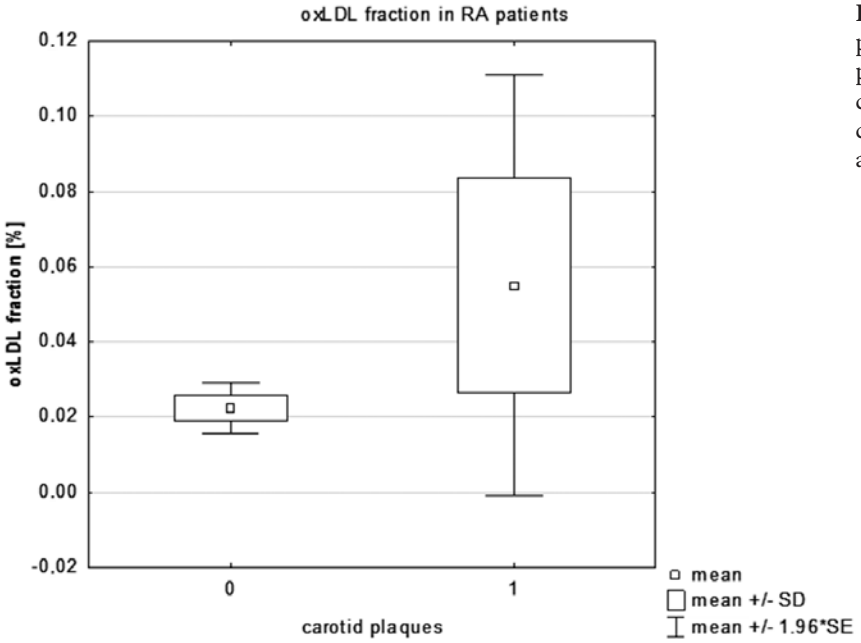

Fig. 1. sOxLDL fraction inRA patients with and without carotid plaques. 0 – patients without carotid plaques, 1 – patients with carotid plaques, RA – rheumatoid arthritis, SD – standard error

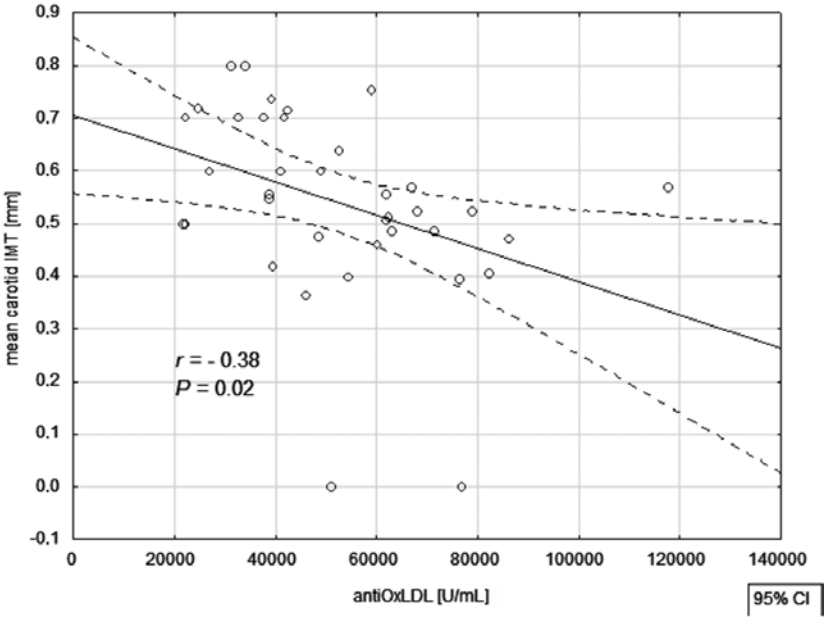

Fig. 2. Correlation between anti--oxLDL level [U/mL] and mean carotid IMT [mm]

In RA patients with carotid plaques, the ox-LDL fraction was significantly higher than in patients without carotid plaques (0.055 ± 0.070 % vs. 0.022 ± 0.018%, p = 0.033) (Fig. 1).

In the RA group, a significant negative correlation was found between the mean carotid IMT and the serum concentration of anti-oxLDL antibodies (RP = –0.38, p = 0.02) (Fig. 2). In addition, in RA patients with thickening of the carotid intima-media complex (IMT > 0.70 mm) the concentration of anti-oxLDL antibodies was significantly lower (37800 ± 1100 U/mL vs. 57600 ± 22300 U/mL, p = 0.009). In RA patients receiving glucocorticoids, a negative correlation was found between the concentration of anti-oxLDL antibodies and the daily prednisone dose (RP = –0.5, p = 0.007).

Discussion

In the present study, no significant differences between the RA group IMT and the control group IMT. This might be caused by the lipid abnormalities in the control group, as significant elevation in LDL-C concentration was noticed in the controls. However, the 10-year risk of fatal CV disease was significantly higher in the RA group. A significantly increased risk of CV events among RA patients has been described by other authors [7].

In the present study, contrary to the reports of other authors, severe thickening of intima-media complex (IMT > 0.90 mm) and/or plaques were detected only in 21.6% of the RA patients. The reason for this discrepancy may be the fact that RA

patients were enrolled in this study without clinical manifestation of atherosclerosis. Additionally, the cohort seems to be younger than groups analyzed in other studies [25, 26]. A younger RA group might also be responsible for the lower mean IMT than in other reports [25]. Although Ahmed et al. analyzed an equally young group of RA patients, severe atherosclerosis was more frequent in their cohort [27]. This might be due to the fact that in 12.5% of the RA patients assessed by Ahmed et al. were diabetic, as opposed to none in the present study; and in 22.5% of the RA patients in those authors' study reported a significant family history of CV diseases, while patients with a significant family history of CVD were excluded from the present study.

As expected, age, a known CV risk factor, was strongly associated with atherosclerosis in both the RA patients and the controls. The data from the study participants is similar in this respect to the general world population.

In the present study, thickening of the intima-media complex and increased SCORE results were observed in anti-CCP-positive RA patients. These results support the thesis that anti-CCP antibodies are among the non-classical CV risk factors in RA patients [5]. Cambridge et al. reported that anti-CCP antibodies might increase the risk of coronary heart disease even in patients without RA [28].

In the RA group in the present study there were associations between the 10-year risk of fatal CV disease (SCORE) and inflammation (ESR), disease activity (DAS28) and disease duration. Ahmad et al. also reported associations between subclinical atherosclerosis and inflammation, disease duration and the number of involved joint areas [29]. In the present study, in RA patients with low disease activity or remission the SCORE results were significantly lower than in patients with moderate to high disease activity. This supports the thesis that the best way to reduce CVD occurrence in RA patients is RA treatment leading to disease control and remission [5].

In RA rheumatoid nodules are among the clinical manifestations of more aggressive disease, but the present study did not find any association between the presence of rheumatoid nodules and ultrasonographic evidence of carotid atherosclerosis (IMT or plaques) or CV risk (according to the modified SCORE). Similar results have been reported by Galarza-Delgado et al. [26].

In the present study a higher fraction of oxLDL was detected in the RA patients than in the controls, and in the RA patients with carotid plaques compared to those without plaques. Ajeganova et al. reported higher oxLDL concentrations in RA patients who experienced a subsequent CV disease [30]. Elevated oxLDL may link chronic inflammation in RA patients with acceleration of atherosclerosis and an increased risk of CV disease in this population. The negative correlation between the concentration of anti-oxLDL antibodies and IMT suggest that these antibodies may be protective and reduce the risk of CVD. Some authors have reported an inverse correlation between IMT and anti-oxLDL antibodies in healthy subjects without any CVD [31]. However, the role of anti-oxLDL antibodies in the development of atherosclerotic changes needs further research. The current authors plan to further investigate the relationship between anti-oxLDL antibodies and CV complications in RA patients by increasing the size of the RA group and following them up.

The present study demonstrated that in RA patients disease activity, ESR, disease duration, the presence of anti-CCP antibodies, a high oxLDL fraction and a low level of anti-oxLDL antibodies may influence the risk of CVD. No influence of rheumatoid nodules on the risk of CVD was detected.

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References

[1] **Gabriel SE:** Why do people with rheumatoid arthritis still die prematurely? Ann Rheum Dis 2008, 67, Suppl 3, 30–34.

[2] **Gonzalez A, Maradit Kremers H, Crowson CS, Nicola PJ, Davis JM3, Therneau TM, Roger VL, Gabriel SE:** The widening mortality gap between rheumatoid arthritis patients and the general population. Arthritis Rheum 2007, 56, 3583–3587.

[3] **Gabriel SE:** Cardiovascular morbidity and mortality in rheumatoid arthritis. Am J Med 2008, 121, 9–14.

[4] **Aviña-Zubieta JA, Choi HK, Sadatsafavi M, Etminan M, Esdaile JM, Lacaille D:** Risk of cardiovascular mortality in patients with rheumatoid arthritis: a meta-analysis of observational studies. Arthritis Rheum 2008, 59, 1690–1697.

[5] **Peters MJL, Symmons DPM, McCarey D, Dijkmans BAC, Nicola P, Kvien TK, McInnes IB, Haentzschel H, Gonzalez-Gay MA, Provan S, Semb A, Sidiropoulos P, Kitas G, Smulders YM, Soubrier M, Szekanecz Z, Sattar N, Nurmohamed MT:** EULAR evidence-based recommendations for cardiovascular risk management in patients with rheumatoid arthritis and other forms of inflammatory arthritis. Ann Rheum Dis 2010, 69, 325–331.

[6] **van Halm VP, Peters MJL, Voskuyl AE, Boers M, Lems WF, Visser M, Stehouwer CDA, Spijkerman AMW, Dekker JM, Nijpels G, Heine RJ, Bouter LM, Smulders YM, Dijkmans BAC, Nurmohamed MT:** Rheumatoid arthritis versus diabetes as a risk factor for cardiovascular disease: a cross-sectional study, the CARRE Investigation. Ann Rheum Dis 2009, 68, 1395–1400.
[7] **del Rincón ID, Williams K, Stern MP, Freeman GL, Escalante A:** High incidence of cardiovascular events in a rheumatoid arthritis cohort not explained by traditional cardiac risk factors. Arthritis Rheum 2001, 44, 2737–2745.
[8] **Crowson CS, Matteson EL, Roger VL, Therneau TM, Gabriel SE:** Usefulness of risk scores to estimate the risk of cardiovascular disease in patients with rheumatoid arthritis. Am J Cardiol 2012, 110, 420–424.
[9] **Graf J, Scherzer R, Grunfeld C, Imboden J:** Levels of C-reactive protein associated with high and very high cardiovascular risk are prevalent in patients with rheumatoid arthritis. PLoS One 2009, 4, e6242.
[10] **Goodson NJ, Farragher TM, Symmons DPM:** Rheumatoid factor, smoking, and disease severity: associations with mortality in rheumatoid arthritis. J Rheumatol 2008, 35, 945–949.
[11] **Sattar N, McCarey DW, Capell H, McInnes IB:** Explaining how "high-grade" systemic inflammation accelerates vascular risk in rheumatoid arthritis. Circulation 2003, 108, 2957–2963.
[12] **Park Y, Ahn C, Choi HK, Lee S, In B, Lee H, Nam C, Lee S:** Atherosclerosis in rheumatoid arthritis: morphologic evidence obtained by carotid ultrasound. Arthritis Rheum 2002, 46, 1714–1719.
[13] **Klop B, Proctor SD, Mamo JC, Botham KM, Castro Cabezas M:** Understanding postprandial inflammation and its relationship to lifestyle behaviour and metabolic diseases. Int J Vasc Med 2012, 2012, 947417.
[14] **Virella G, Virella I, Leman RB, Pryor MB, Lopes-Virella MF:** Anti-oxidized low-density lipoprotein antibodies in patients with coronary heart disease and normal healthy volunteers. Int J Clin Lab Res 1993, 23, 95–101.
[15] **Nowak B, Szmyrka-Kaczmarek M, Durazińska A, Płaksej R, Borysewicz K, Korman L, Wiland P:** Anti-ox-LDL antibodies and anti-ox-LDL-B2GPI antibodies in patients with systemic lupus erythematosus. Adv Clin Exp Med 2012, 21, 331–335.
[16] **Inoue T, Uchida T, Kamishirado H, Takayanagi K, Hayashi T, Morooka S:** Clinical significance of antibody against oxidized low-density lipoprotein in patients with atherosclerotic coronary artery disease. J Am Coll Cardiol 2001, 37, 775–779.
[17] **Karvonen J, Päivänsalo M, Kesäniemi YA, Hörkkö S:** Immunoglobulin M type of autoantibodies to oxidized low-density lipoprotein has an inverse relation to carotid artery atherosclerosis. Circulation 2003, 108, 2107–2112.
[18] **Deanfield JE, Halcox JP, Rabelink TJ:** Endothelial function and dysfunction: testing and clinical relevance. Circulation 2007, 115, 1285–1295.
[19] **van den Oord SCH, Sijbrands EJG, ten Kate GL, van Klaveren D, van Domburg RT, van der Steen AFW, Schinkel AFL:** Carotid intima-media thickness for cardiovascular risk assessment: systematic review and meta-analysis. Atherosclerosis 2013, 228, 1–11.
[20] **Evans MR, Escalante A, Battafarano DF, Freeman GL, O'Leary DH, del Rincón I:** Carotid atherosclerosis predicts incident acute coronary syndromes in rheumatoid arthritis. Arthritis Rheum 2011, 63, 1211–1220.
[21] **Conroy RM, Pyörälä K, Fitzgerald AP, Sans S, Menotti A, De Backer G, De Bacquer D, Ducimetière P, Jousilahti P, Keil U, Njølstad I, Oganov RG, Thomsen T, Tunstall-Pedoe H, Tverdal A, Wedel H, Whincup P, Wilhelmsen L, Graham IM:** Estimation of ten-year risk of fatal cardiovascular disease in Europe: the SCORE project. Eur Heart J 2003, 24, 987–1003.
[22] **Touboul PJ, Hennerici MG, Meairs S, Adams H, Amarenco P, Bornstein N, Csiba L, Desvarieux M, Ebrahim S, Fatar M, Hernandez Hernandez R, Jaff M, Kownator S, Prati P, Rundek T, Sitzer M, Schminke U, Tardif JC, Taylor A, Vicaut E, Woo KS, Zannad F, Zureik M:** Mannheim carotid intima-media thickness consensus (2004–2006). An update on behalf of the Advisory Board of the 3rd and 4th Watching the Risk Symposium, 13th and 15th European Stroke Conferences, Mannheim, Germany, 2004, and Brussels, Belgium 2006.
[23] **Salonen JT, Salonen R:** Ultrasound B-mode imaging in observational studies of atherosclerotic progression. Circulation 1993, 87, 56–65.
[24] **Ikeda N, Kogame N, Iijima R, Nakamura M, Sugi K:** Carotid artery intima-media thickness and plaque score can predict the SYNTAX score. Eur Heart J 2012, 33, 113–119.
[25] **Corrales A, Parra JA, González-Juanatey C, Rueda-Gotor J, Blanco R, Llorca J, González-Gay MA:** Cardiovascular risk stratification in rheumatic diseases: carotid ultrasound is more sensitive than Coronary Artery Calcification Score to detect subclinical atherosclerosis in patients with rheumatoid arthritis. Ann Rheum Dis 2013, 72, 1764–1770.
[26] **Galarza-Delgado DA, Esquivel-Valerio JA, Garza-Elizondo MA, Góngora-Rivera F, Muñoz-De Hoyos JL, Serna-Peña G:** Carotid atherosclerosis in patients with rheumatoid arthritis and rheumatoid nodules. Reumatol Clin 2013, 9, 136–141.
[27] **Ahmed HMMS, Youssef M, Mosaad YM:** Antibodies against oxidized low-density lipoprotein are associated with subclinical atherosclerosis in recent-onset rheumatoid arthritis. Clin Rheum 2010, 29, 1237–1243.
[28] **Cambridge G, Acharya J, Cooper JA, Edwards JC, Humphries SE:** Antibodies to citrullinated peptides and risk of coronary heart disease. Atherosclerosis 2013, 228, 243–246.
[29] **Ahmad S, Garg S, Dhar M, Srivastava S, Biswas D, Barthwal SP, Shirazi N, Srivastava R:** Predictors of atherosclerosis in rheumatoid arthritis. Vasa 2012, 41, 353–359.
[30] **Ajeganova S, de Faire U, Jogestrand T, Frostegård J, Hafström I:** Carotid atherosclerosis, disease measures, oxidized low-density lipoproteins, and atheroprotective natural antibodies for cardiovascular disease in early rheumatoid arthritis – an inception cohort study. J Rheumatol 2012, 39, 1146–1154.
[31] **Fukumoto M, Shoji T, Emoto M, Kawagishi T, Okuno Y, Nishizawa Y:** Antibodies against oxidized LDL and carotid artery intima-media thickness in a healthy population. Arterioscler Thromb Vasc Biol 2000, 20, 703–707.

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