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Rheumatoid arthritis as a vascular disease

by György Kerekes

Supervisor: Pál Soltész MD. PhD.

UNIVERSITY OF DEBRECEN
KÁLMÁN LAKI DOCTORAL SCHOOL OF THROMBOSIS, HEMOSTATIS,
AND VASCULAR DISEASES

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Impressum

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by György Kerekes, M.D.

Supervisor: Pál Soltész, D.Sc.

Kálmán Laki Doctoral School of Thrombosis, Hemostatis, and Vascular Diseases
University of Debrecen

Head of the **Examination Committee**:	Dr. László Muszbek, M.H.A.Sc
Members of the Examination Committee:	Dr. József Balla, D.Sc. Dr. László Kovács, Ph.D.

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Head of the **Defense Committee**:	László Muszbek, M.H.A.Sc
Reviewers:	Dr. László Entz, Ph.D. Dr. János Gaál, Ph.D.
Members of the Defense Committee:	Dr. József Balla, D.Sc. Dr. László Kovács, Ph.D.

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1. Introduction

Rheumatoid arthritis (RA) is a chronic inflammatory disease eventually leading to joint destruction, impaired articular function, and physical disability. With the introduction of novel therapies and better patient care, the quality of life in RA has significantly improved during the past years without improvement of life expectancy. Thus, chronic comorbidities including vascular diseases and malignancies in association with RA have become major issues. Among mortality factors, amyloidosis and infection have been replaced by cardiovascular disease (CVD) which is now the leading cause of death in RA patients. Recently, the importance of accelerated atherosclerosis and high risk for CVD has been acknowledged as main factors causing reduced life expectancy by 5–10 years in RA. In addition, there is increasing body of evidence suggesting that increased cardiovascular (CV) risk in RA is comparable to that of type 2 diabetes mellitus.

Both RA and atherosclerosis are associated with chronic inflammation, and RA is an independent risk factor for accelerated atherosclerosis. Both classical CV, as well as systemic inflammation-associated risk factors have been implicated in accelerated atherosclerosis and vascular disease in RA. Numerous recent studies have demonstrated the role of classical, Framingham, and inflammation-associated risk factors in this context. Regarding traditional risk factors, cigarette smoking has clearly been implicated in the development of atherosclerosis in RA. Smoking has also been associated with seropositive RA itself and may exert a relationship with disease severity as well. Data on dyslipidemia in RA are conflicting, and decreased plasma high-density lipoprotein (HDL-C) and increased low-density lipoprotein cholesterol (LDL-C) levels may be secondary to systemic inflammation. There is no clear evidence that hypertension, diabetes mellitus, obesity, and sedentary lifestyle are directly implicated in accelerated atherosclerosis in RA.

As RA-associated atherosclerosis cannot be solely explained by Framingham risk factors, the inflammatory mechanisms underlying RA may be crucial for early atherosclerosis and CVD development. Atherosclerotic plaque and RA synovial tissue are in many ways similar to each other as they both contain inflammatory leukocytes, mainly T cells and macrophages, pro-inflammatory cytokines including tumor necrosis factor-α (TNF-α), chemokines, heat shock proteins, matrix-degrading enzymes, endothelial adhesion molecules and several other inflammatory mediators. Endothelial activation and dysfunction as the first step of atherogenesis have been associated with the increased expression of endothelial adhesion molecules. It has been shown that sustained inflammatory activity may be the predominant risk factor for accelerated atherosclerosis and excess CVD mortality in RA.

Nowadays there are some noninvasive tests which are able to detect early signs of atherosclerosis in clinical and experimental circumstances. Endothelial dysfunction measured by duplex ultrasound on the brachial artery is the earliest, sensitive and reproducible marker of accelerated atherosclerosis in vivo. The flow mediated vasodilation (FMD) predicts the presence of coronary artery disease (CAD) and the short term cardiovascular events during vascular surgery. Carotid intima-media thickness (ccIMT) is a novel indirect measure of total atherosclerotic burden. The ccIMT has a good predictive value to determine future cardiovascular events. Measurement of ccIMT as a well standardized parameter is a useful method for defining the true cardiovascular risk in patients with intermediate cardiovascular risk in clinical practice.

2. Aims

1. First we assessed brachial FMD as early indicator of endothelial dysfunction, ccIMT, a marker of atherosclerosis, as well as laboratory markers of inflammation, autoimmunity, and accelerated atherosclerosis in Hungarian RA population. We wished to determine the difference of total atherosclerotic burden between RA and control subjects and to find correlation between the parameters of systemic inflammation and atherosclerotic measures.

2. In the second study, we followed the signs of endothelial function, ccIMT and levels of lipid parameters before and after rituximab therapy. Our goal was to determine the effect of B cell depletion therapy on parameters of subclinical atherosclerosis and lipid profile in RA patients.

3. The severity and outcome of CVD may also depend on disease duration in RA as atherosclerosis becomes more evident with the progression of the disease. Most of the studies on vascular dysfunction, atherosclerosis and CVD were performed in long-lasting established RA. In the third study we measured the laboratory and ultrasound markers of subclinical atherosclerosis and disease activity during adalimumab treatment in early RA patients. We wanted to assess the potential reversibility of atherosclerotic changes in early stage of disease course.

3. Patients and methods

3.1. First study

3.1.1. Patients and controls

Fifty-two patients with RA (40 women, 77%, and 12 men, 23%; mean age 51 ± 12 yrs, range 23–77 yrs; all Caucasian) and 40 age- and sex matched healthy control subjects (31 women, 77.5%, and 9 men, 22.5%; mean age 50 ± 10 yrs, range 26–76 yrs; all Caucasian) were included in our study. All patients with RA fulfilled the American College of Rheumatology (ACR) criteria for RA. The mean disease duration of RA was 10.5 ± 8.5 years (range 2–34 yrs). Exclusion criteria included known cardiovascular and cerebrovascular diseases, hypertension (blood pressure > 140/90 mm Hg), diabetes mellitus, cigarette smoking, obesity [body mass index (BMI) ≥ 30 kg/m^2], rheumatoid vasculitis, current infectious disease, or renal failure (serum creatinine ≥ 117 mmol/l). All patients and controls were fasting and had not used alcohol, tobacco, antioxidants, and vasoactive drugs within the past 24 hours. No patient received corticosteroids at the time of and at least 3 months prior to the study in order to exclude the atherogenic effects of these compounds. The control subjects were recruited from volunteering hospital staff members and visitors in an age- and sex-matched manner. Patients with RA and controls were also normalized for Framingham traditional risk factors for atherosclerosis.

3.1.2. Methods

Physical and laboratory examinations

Demographic data including sex, age, and disease duration were recorded at the time of the study. Thorough examinations including assessment of height, weight, blood pressure,

and calculation of BMI were performed. After overnight fasting, blood samples were collected from the patients and controls for serum glucose, total cholesterol (TC), low density lipoprotein-cholesterol (LDL-C), high-density lipoprotein (HDL-C), triglyceride (Tg) levels, renal and liver function tests, and full blood count.

Regarding immunological markers, serum IgM RF and CRP were assessed by quantitative nephelometry (Cobas Mira Plus, Roche), using RF and CRP reagents, respectively (both Dialab, Vienna, Austria). RF levels > 50 IU/ml and high sensitivity CRP levels > 5 mg/l were considered elevated. Anti-CCP autoantibodies were detected in serum samples using the Immunoscan-RA CCP2 ELISA test (Euro-Diagnostica, Arnhem, The Netherlands). The assay was performed according to the instructions of the manufacturer. A concentration > 25 IU/ml was considered positive. Serum total immunoglobulin A (IgA), IgG, and IgM levels were assessed by turbidimetry (Dialab). ANA were screened on HEp-2 cells by immunofluorescence. Serum levels of anti-dsDNA autoantibodies were determined by an EIA kit (BioSystems, Barcelona, Spain). Total complement hemolytic activity of the classical pathway was measured by the CH50 assay, adapted on a 96-well microplate, with results expressed in hemolytic units. Complement C3 and C4 serum levels were measured by nephelometry (Behring, Marburg, Germany). Circulating immune complexes (CIC) in the sera were assessed by polyethylene glycol (PEG) precipitation. For detection of anti-ß2-GPI and anti-CL antibodies, commercial ELISA systems were used (Orgentec, Mainz, Germany). Quantification of anti-oxLDL antibodies was performed by a commercial enzyme immunoassay (Immco, Buffalo, NY, USA) according to the manufacturer's instructions. Regarding cytokines, serum TNF-α, IL-1, IL-4, IL-6, IL-10, IFN-γ, and transforming growth factor-ß (TGF-ß) levels were determined by ELISA (R & D Systems). Total lymphocyte counts and lymphocyte subsets including the percentage and absolute numbers of CD3+, CD4+, CD8+, CD19+, andCD56+ T cell subsets were also determined using antigen-specific

monoclonal antibodies (all Sigma). The percentage and absolute numbers of Th0 (CD4+/IFN-γ+/IL-4+), Th1 (CD4+/IFN-γ+/IL-4–), Th2 (CD4+/IFN-γ–/IL-4+), Tc0 (CD8+/IFN-γ+/IL-4+), Tc1 (CD8+/IFN-γ+/IL-4–), and Tc2 (CD8+/IFN-γ–/IL-4+) subpopulations were also assessed. Anti-human IFN-γ-FITC and anti-human IL-4-PE antibodies were purchased from Becton-Dickinson.

Ultrasound diagnostic of subclinical atherosclerosis

Brachial flow mediated vasodilation (FMD) and nitrate mediated vasodilation (NMD) were carried out to estimate the vascular function. Ultrasound examination was performed on the right arm using a 10 MHz linear array transducer (HP Sonos 5500 ultrasound system) by a single trained sonographer after 30 min resting in a temperature-controlled room (basal value for FMD). A B-mode longitudinal section was obtained of the brachial artery above the antecubital fossa. In order to assess FMD, reactive hyperemia was induced by release of a pneumatic cuff around the forearm inflated to suprasystolic pressure for 4.5 min. After deflation the maximal flow velocity and arterial diameter was continuously recorded for 90 s. After 15 min of recovery to the baseline diameter (basal value for NMD), 400 μg sublingual nitroglycerine was administered and NMD was assessed. Flow velocities, baseline diameter, FMD, and NMD were electrocardiogram-gated and recorded offline. FMD and NMD values were expressed as percentage change from baseline (resting) value (FMD and NMD).

The ccIMT measurements were carried out by a single observer. The same duplex ultrasound system (HP Sonos 5500, 10 MHz linear array transducer) was used to assess the common carotid arteries. Longitudinal high-resolution B-mode ultrasound scans were employed over both right and left common carotid arteries and were R-synchronized and recorded. The offline measurements were performed 1 cm proximal to the carotid bulb in the far wall. The ccIMT was defined as the distance between the first and second echogenic lines

from the lumen, taking the average of 10 measurements on both sides. ccIMT values were expressed in millimetres.

3.2. Second study

3.2.1. Patients

Five female RA patients (mean age: 41.6 years, range: 29-56 years) were included in the study. The mean disease duration was 5.8 years (1-9 years). All patients received 1000 mg i.v. rituximab on March 1, 2007, and a second i.v. infusion two weeks later after 100 mg i.v. methylprednisolone premedication. As RTX is approved as a second-line biologic agent in Hungary, all patients had received at least one TNF blocker. However, there was a wash-out period of at least 3 months before the introduction of RTX. All patients received stable doses (10-20 mg weekly) of oral methotrexate at least 6 months before and throughout the study. We used the same inclusion and exclusion criteria as described above in first study protocol.

3.2.2. Methods

Brachial FMD and ccIMT values were assessed at baseline (before the first infusion), after 2 weeks (before the second infusion) then at week 6 and at week 16 as described above.

Before every vascular examination blood samples were taken from the patients. Serum IgM RF, CRP, anti-CCP, total cholesterol (TC), triglyceride (Tg), LDL-C and HDL-C were assessed as described above.

3.3. Third study

3.3.1. Patients

Our study group comprised eight early RA patients: 6 women and 2 men with a mean age of 37.8 years (range 24–69). The duration of disease was at least 1 year in all patients; the mean duration was 5.6 months (range 3–12 months). None of the patients have ever received methotrexate or any biologics, and none had taken corticosteroids for at least 3 months prior to the study. According to the protocol, all patients received 40 mg adalimumab subcutaneously every 2 weeks. Concomitant methotrexate therapy of 10–15 mg/week was initiated in all patients. We used the same exclusion criteria as described above in first study protocol.

3.3.2. Methods

Since endothelial function may change rather rapidly, FMD values were assessed at baseline (before the first injection) and then after 2, 4, 8 and 12 weeks. ccIMT values were assessed at baseline (before the first infusion) and after 24 weeks, because a relatively longer time is needed to detect changes in carotid atherosclerosis.

Before every vascular examination blood samples were obtained from the patients. Circulating von Willebrand factor (vWF) antigen is a marker of endothelial cell activation. Plasma levels of vWF were determined by STA Liatest vWF immunoturbidimetric assay using microlatex particles coated with polyclonal rabbit anti-human vWF antibodies (Diagnostica Stago, Asnieres, France). After mixing the reagent with plasma, the degree of agglutination was proportional to the amount of vWF present in the plasma sample. The reference range was 50–160%. Serum IgM RF, CRP, anti-CCP, total cholesterol, triglyceride, LDL-C and HDL-C were assessed as well.

3.4. Statistical analysis

The statistical analysis was performed using the SPSS software version 15.0. The descriptive data of normal variables are expressed as the mean ± SD. Statistical analysis was carried out by independent, 2-tailed t-test. Correlations between variables were determined using Pearson correlation analysis for normally distributed values and Spearman correlation analysis as nonparametric test. R values of these correlations were determined and corresponding p values < 0.05 were considered significant. Since there were only five and eight patients in these pilot studies, a limited statistical analysis could have been performed.

Regarding reproducibility, all assessments were performed by a single observer. Intra-observer variability of FMD and ccIMT measurements was manually calculated as 5% and 4.2%, respectively. According to the intraclass correlation coeffecients (FMD-ICC 0,935; ccIMT-ICC 0,976) and Bland-Altman (MedCalc) analysis the reproducibility of both vascular examinations are excellent in our laboratory.

4. Results

4.1. First study

4.1.1. Assessment of FMD, NMD, and ccIMT in RA and control groups

The RA patient group and the control group were matched with regard to traditional Framingham risk factors. FMD in patients with RA expressed as percentage of the basal value was significantly lower (5.32% ± 4.66%) compared to controls (8.30% ± 3.96%) ($p = 0.001$). However, no significant difference was found in NMD between patients with RA (18.30% ± 15.17%) and controls (17.50% ± 6.96%) (NS). The ccIMT was significantly higher in patients (0.63 ± 0.14 mm) compared to controls (0.54 ± 0.15) ($p = 0.012$).

4.1.2. Correlations between FMD, NMD, ccIMT, epidemiological and laboratory markers

Within the RA patient population, FMD, NMD, and ccIMT values were correlated with each other, as well as with other epidemiological and laboratory indicators. We found a significant negative correlation between ccIMT and FMD ($R = –0.318$, $p = 0.022$). The FMD correlated negatively with disease duration independently from age (R=-0,414, p=0,040). ccIMT showed a significant positive correlation with age ($R = 0.831$, $p < 0.001$), serum total cholesterol ($R = 0.285$, $p = 0.041$), TNF-α levels (R=0,321, p=0,038) and a significant inverse correlation with serum IL-1 levels ($R = –0.773$, $p < 0.001$). Interestingly, there was a significant, positive correlation between ccIMT and anti-dsDNA levels ($R = 0.463$, $p = 0.006$), although the absolute value of anti-dsDNA was within the normal range in all patients. FMD% was positively correlated with serum IFN-γ levels ($R = 0.516$, $p = 0.014$) and inversely correlated with total lymphocyte counts ($R = –0.451$, $p = 0.04$).

4.1.3. Comparisons of RA patients with low versus high ccIMT and those with impaired versus normal FMD.

As the whole RA patient population was rather heterogenous regarding FMD and ccIMT, we divided patients with RA into low (< 0.65 mm; n = 26) and high (> 0.65 mm; n = 26) ccIMT groups, and into "normal" (> 5%; n = 27) and "impaired" (< 5%; n = 25) FMD subsets (Table 4). Regarding ccIMT, the low and high groups differed significantly in age (43.7 ± 8.3 versus 58.2 ± 10.1 years; p = 0.001), serum IL-1 (26.8 ± 14.2 vs 6.2 ± 10.9 pg/ml; p = 0.02), anti-dsDNA (8.3 ± 3.9 vs 16.8 ± 12.4 IU/ml; p = 0.011), and IFN-γ levels (21.7 ± 27.3 vs 8.4 ± 11.3 pg/ml; p = 0.04). Regarding FMD, the normal and impaired groups differed significantly in age (45.4 ± 9.8 vs 56.1 ± 11.1 yrs; p = 0.001), disease duration (8.6 ± 5.5 vs 14.4 ± 12.7 yrs; p = 0.042), and serum IFN-γ levels (22.6 ± 28.6 vs 8.8 ± 11 pg/ml; p = 0.037).

4.2. Second study

4.2.1. Effects of rituximab treatment on endothelial dysfunction

To assess endothelial function, brachial artery FMD was measured by high resolution ultrasonography. The baseline FMD values of the five patients were 2.1%, 4.2%, 2.9%, 4.5%, 5.9%,, which values improved by week 16 to 4.0%, 8.9%, 3.6%, 8.4% and 11.4%, respectively. FMD improved in 4/5 patients by week 2, right before the administration of the second infusion and FMD increased in all patients by week 16. When comparing FMD values at weeks 2, 6 and 16 to the baseline (week 0), the mean FMD improvement was 29.9% at week 2, 21.6% at week 6 and 80.8% at week 16. This improvement was statistically significant in comparison to the baseline values (3.92±1.47 vs. 7.24±3.35, p=0.02).

4.2.2. Effects of rituximab treatment on lipid profile

Plasma total cholesterol (TC), HDL-C, LDC-C and triglyceride (Tg) were assessed at baseline, at weeks 2, 6 and 16 in all five patients. Initially, TC levels were between 3.9-5.8 mmol/l, HDL-C between 0.8-1.3 mmol/l, LDL-C between 1.8-3.8 mmol/l and Tg between 0.7-1.3 mmol/l. Percentages of changes in comparison to the baseline levels (ΔTC, ΔHDL-C, ΔLDL-C and ΔTg) were calculated at weeks 2, 6 and 16. TC levels decreased in 4/5 patients by a mean 2.6% at week 2, 10.8% at week 6 and 8.5% at week 16. Mean HDL-C production increased by 14.3%, 33.1% and 35.4%, respectively, the improvement was statistically significant (1.49±0.35 vs. 2.05±0.74, p=0.035). LDL-C concentrations eventually decreased in two and increased in 3 patients resulting in a mean 2.3-3.6% decrease over the observation period. Finally, Tg variably decreased or increased in the patients, the net effect by week 16 was around zero.

4.3. Third study

4.3.1. Clinical response of patients to adalimumab

After 12 weeks of adalimumab therapy, CRP levels significantly decreased from 52.8 ± 36.1 to 8.3 ± 5.0 mg/L (p=0.04). In addition, DAS28 also significantly decreased from 5.98 ± 0.76 to 2.54 ± 0.53 (p=0.0001). Anti-CCP and RF concentrations did not change with adalimumab therapy.

4.3.2. Effect of adalimumab treatment on endothelial function and ccIMT

FMD improved in all patients as early as week 2. The mean absolute values of FMD at baseline and at weeks 2, 4, 8 and 12 were 7.0 ± 5.9%, 10.6 ± 3.2%, 11.1 ± 4.2%, 11.9 ± 5.1% and 13.2 ± 5.6%, respectively. This indicated 51.4%, 58.6%, 70.0% and 88.6%, all significant

improvements at weeks 2, 4, 8 and 12 in comparison to baseline, respectively ($p<0.05$ at all time points).

Although atherosclerosis may be a more prolonged process in comparison to endothelial dysfunction and improvement in ccIMT cannot be expected in the short term, when assessing ccIMT by ultrasonography we could detect notable changes after 24 weeks of treatment in comparison to baseline. The ccIMT values at baseline and week 24 were 0.59 ± 0.09 mm and 0.52 ± 0.06 mm, respectively. This indicated an 11.9% significant improvement at week 24 in comparison to baseline ($p=0.002$).

4.3.3. Effect of adalimumab treatment on vWF level

We assessed circulating vWF levels as indicators of endothelial activation. Plasma vWF decreased in 5/8 patients by as early as week 2. By the 12th week, plasma vVW levels decreased in 6 from 8 patients. The mean± SD plasma vWF levels at baseline and weeks 2, 4, 8 and 12 were 225.8 ± 89.3%, 225.3 ± 79.5%, 197.4 ± 66.5%, 185.6 ± 66.0% and 184.8 ± 60.3%, respectively. This indicated 0.2%, 12.6%, 17.8% and 18.2% decreases at weeks 2, 4, 8 and 12 in comparison to baseline, respectively, but these changes did not reach statistical significance.

4.3.4. Correlation between vascular and laboratory parameters

There was a significant inverse correlation between FMD and CRP ($R=-0.596$, $p=0.015$). Although, as presented above, only non-significant decrease in plasma vWF levels could be observed during adalimumab treatment, we also found a negative correlation between FMD and plasma vWF ($R=-0.643$, $p=0.007$). Interestingly, CRP and vWF also correlated with each other ($R=0.598$, $p=0.014$). These results indicate that endothelial dysfunction may be associated with systemic inflammation, as well as endothelial activation.

5. Discussion

5.1. First study

In our cross sectional controlled study we assessed brachial artery flow mediated dilation (FMD) as indicator of endothelial dysfunction, common carotid intima-media thickness (ccIMT), an early marker of atherosclerosis, as well as laboratory markers of inflammation, autoimmunity and accelerated atherosclerosis in RA and control population.

Even after excluding the influence of traditional risk factors (cigarette smoking, hypertension, diabetes, dyslipidemia, obesity) we still found endothelial dysfunction and progressive atherosclerosis in RA patient indicating the potential role of inflammation in the atherosclerotic process. We observed positive correlation between endothelial dysfunction (as the first step of atherosclerosis) and disease duration (as a disease related factor) independently on age. Our results and several other studies demonstrate that the pathogenesis of RA and atherosclerosis may overlap. Although RA-associated atherosclerosis also involves traditional Framingham risk factors, such as cigarette smoking, hypertension, diabetes, dyslipidemia, or obesity, these do not fully account for the development of vascular damage in RA.

Experimental data suggest that the inflammation in atherosclerosis is based on a Th1 type immune mechanism. Interestingly, the typical Th1 type cytokines such as IL-1 and interferon-γ were inversely associated with early signs of atherosclerosis that support the need for further studies to shed light on the details of the molecular mechanisms of inflammation associated with atherosclerosis.

Anti-dsDNA levels showed strong positive correlation with ccIMT. Anti-dsDNA antibodies can be detected in patients with RA, but the serum anti-dsDNA levels in patients with RA remained within the normal range. However, patients with SLE and non

differentiated connective tissue disease also experience accelerated atherosclerosis in connection with anti-dsDNA levels. Anti-dsDNA may be a serum marker of generalized atherosclerosis or may participate in the pathogenesis of inflammation induced atherosclerosis.

5.2. Second study

In our first pilot study we assessed the effects of rituximab (anti-CD20 antibody blocking the B cell function) on FMD, ccIMT and lipid profile during a 16-week follow up. Rituximab treatment resulted in a rapid and sustained improvement in FMD without significant decrease in ccIMT. Rituximab exerted early and sustained favorable effects on plasma total cholesterol and HDL-C levels.

There have been no reports regarding the influence of rituximab treatment on vascular function and atherosclerosis in RA before our publication. Our results indicated that the potential beneficial effect of B cell depletion therapy on endothelial dysfunction depends on the control of disease activity and the improvement of metabolic state as well.

5.3. Third study

Several reports indicated that TNF-α blockers may exert favorable but transient effects on FMD of the brachial artery and ccIMT in RA. In our pilot study we investigated the vascular effects of adalimumab (a humanized TNF-α blocker) on disease activity in recent onset RA. Adalimumab therapy considerably improved arthritis as it decreased CRP levels and disease activity (DAS28), and resulted in a significant increase in FMD by as early as week 2. Significantly, these effects were sustained until week 12. Furthermore, the production of vWF, a marker of endothelial activation was also decreased. Regarding carotid

atherosclerosis, after 24 weeks of adalimumab treatment a significant improvement in ccIMT was observed.

In conclusion, blocking the typical Th1 type cytokine (TNF-α) with adalimumab improved endothelial function and postponed the development of atherosclerosis in strong correlation with disease activity in early RA.

Our results suggest that effective inhibition of the inflammatory process carried out both with TNF-α blocking agents or with B cell depletion seems to be beneficial for endothelial function to delay early atherosclerosis. These effects can be partly explained by direct anti-inflammatory potential of these agents besides their impact on classical risk factors (e.g. lipid homeostasis).

6. Summary – New results

1. We were among the first to examine the early signs of atherosclerosis and endothelial dysfunction simultaneously in RA patients in connection with cytokine profile.
2. We have found elevated ccIMT and decreased FMD values in a Hungarian RA population as signs of accelerated atherosclerosis.
3. We have indirectly verified the potential role of inflammation in atherosclerosis regarding RA.
4. Our study demonstrated an age independent harmful effect of long disease duration to endothelial function in RA.
5. We were the first ones to show the bimodal effects of typical Th1 cytokines on the development of atherosclerosis in RA.
6. We found a strong positive correlation between ccIMT and anti-dsDNA level. The latter one may be a marker of accelerated atherosclerosis in RA in the future.
7. We were the first to examine the effect of adalimumab treatment on endothelial function and ccIMT in early RA.
8. Our results indicated the importance of early effective treatment in patients with recent onset RA. Adalimumab administration resulted in striking and long lasting improvement of endothelial function and ccIMT parallel with clinical remission.
9. We were the first to examine the vascular effect of rituximab treatment in resistant RA.
10. B cell depletion therapy exerted a beneficial effect on endothelial dysfunction in RA which partly related on a favourable change in lipid metabolism.

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List of publications related to the dissertation

1. **Kerekes, G.**, Soltész, P., Szűcs, G., Szamosi, S., Dér, H., Szabó, Z., Csáthy, L., Váncsa, A., Szodoray, P., Szegedi, G., Szekanecz, Z.: Effect of Adalimumab Treatment on Vascular Disease Associated with Early Rheumatoid Arthritis.
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