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Impairment of endothelial function in patients with multiple sclerosis

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Abstract

OBJECTIVE: Large epidemiological studies suggest higher risk of vascular events in patients with multiple sclerosis (MS). Chronic inflammatory response and oxidative stress, key-players in a process of atherogenesis, are also suspected to play a role in pathophysiology of MS. Prospective studies elucidating risk of atherosclerosis in MS patients are currently missing. The aim of the study was to assess endothelial function in patients with MS and in healthy controls.
METHODS: We enrolled 46 patients with diagnosis of relapsing-remitting MS and age-matched population of 31 healthy subjects. Endothelial function was assessed using peripheral arterial tonometry and expressed as reperfusion hyperemia index (RHI).
RESULTS: RHI in MS population was significantly lower than in controls (1.77 vs 2.30; p=0.001), even though control population seemed to have higher burden of known vascular risk factors (significantly higher portion of male sex and significantly higher body mass index; $p \leq 0.001$ for both parameters). The presence of MS was the only significant independent variable associated with the RHI (beta=0.396, $p<0.001$) in multiple linear regression model.
CONCLUSION: Results of our study suggest significant impairment of endothelial function in MS population compared to age matched control population with low burden of vascular risk factors.

INTRODUCTION

Multiple sclerosis (MS) is believed to be an immune-mediated neurodegenerative disorder of the human central nervous system (CNS), which usually affects younger adults with certain genetic background (Tomalka-Kochanowska *et al.* 2014). The etiology of MS is unclear, but the most widely accepted hypothesis for the pathogenesis of MS is an autoimmune process leading to neuroinflammation and demyelination. Antimyelin T-cell-mediated inflammatory responses are believed to have a crucial role in the development of focal lesions. Chronic inflammatory response and oxidative stress are also partly involved in the pathophysiology of MS (Weiner 2004; Frohman *et al.* 2006). These pathologic mechanisms are also associated with endothelial dysfunction. Alterations in endothelial function are believed to be one of the most important initial steps in the atherogenesis

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(Siarnik *et al.* 2014). Increase in some laboratory markers of endothelial dysfunction have also been reported in MS patients (Minagar *et al.* 2001). Findings from epidemiological studies suggest, that patients with MS have a higher risk for ischemic stroke and other cardiovascular events, than population without MS (Allen *et al.* 2008; Christiansen *et al.* 2010). Atherosclerosis is one of the key mechanisms responsible for cardiovascular events. The aim of this study was to compare the values of non-invasively assessed endothelial function in patients with MS and in healthy controls.

MATERIAL AND METHODS

The MS population was recruited from the patients followed-up at the Center for Multiple Sclerosis, 1st Department of Neurology, Faculty of Medicine, Comenius University, Bratislava. Study was approved by the institutional ethics committee and all patients provided informed consent.

We enrolled 46 patients with clinically defined relapsing-remitting MS (mean age 37.26 ± 9.50 years) and age-matched population of 31 healthy subjects with a low burden of vascular risk factors (mean age 38.81 ± 10.12). Exclusion criteria for both populations included internal diseases, current use of intravenous corticosteroids and chronic medication except of current disease-modifying treatment (DMT). All clinical characteristics were scored at the time of endothelial function assessment.

Assessment of endothelial function

Reperfusion hyperemia index (RHI), which is a parameter of endothelial function, was assessed non-invasively using the EndoPAT 2000 device (Itamar Medical, Caesarea, Israel). The measurement was calculated using a computerized automated algorithm (software version 3.1.2) provided with the device. Measurements were performed according to the developer´s instructions. Briefly, the subjects were in supine position for a minimum of 20 minutes before the measurements, in a quiet, temperature-controlled (21–24 °C) room with dimmed lights. The subjects were asked to remain perfectly still and silent during the entire measurement period. Each recording consisted of 5 minutes of baseline measurement, 5 minutes of occlusion measurement, and 5 minutes of post-occlusion measurement (hyperemic period). Occlusion of the brachial artery was performed on the non-dominant upper arm. The occlusion pressure was at least 60 mmHg above the systolic blood pressure. The RHI measure of ≤1.67 was considered as endothelial dysfunction (Axtell *et al.* 2010).

Statistical analysis

The analyses were assessed with SPSS version 18 (SPSS Inc., Chicago, USA). Categorical variables were expressed as numbers and proportions (%). Continuous variables were expressed as means ± standard deviation or median, interquartile range, range. To determine relationships between RHI and population characteristics, Pearson or Spearman correlation coefficients were used. Stepwise multiple regression analysis was used to identify predictors of RHI. To compare group, chi-squared test, Student's t-test or Mann-Whitney test were used for particular variables. All tests were 2 sided, p-values <0.05 were considered statistically significant.

RESULTS

Characteristics of study populations are included in Table 1. In MS population, the median duration of the disease was 84 (131.3, 3.0–336.0) months, mean Expanded Disability Status Scale (EDSS) was 2.34 ± 1.1 and current attack of the disease was present in 13 (28.3%) patients. 16 patients (34.8%) were currently taking no DMT, 10 patients (21.7%) were taking interferone beta-1b, 16 patients (34.8%) glatirameracetate and 4 patients (8.7%) natalizumab.

The values of endothelial function in the MS population were significantly lower than in control group (1.77, 0.71, 0.46–2.98 vs 2.30, 0.98, 1.58–3.43; p=0.001). Control group consisted of subjects with a significantly higher proportion of males (17.39% vs 77.42%; p≤0.001), with significantly higher average body mass index (BMI) values (22.83 ± 4.51 vs 28.20 ± 2.93; p≤0.001) and non-significantly higher proportion of smokers (10.87% vs 25.81%; p=0.086).

We failed to find any significant correlation among the values of RHI and disease characteristics, such as EDSS (r=–0.103, p=0.496) and disease duration (r=0.032, p=0.831). There was no difference in RHI in populations with and without current attack of the disease (p=0.942). RHI was significantly higher in patients treated with glatirameracetate than in patients treated with interferone beta-1b (p=0.023) and those without any DMT (p=0.011), as shown in Figure 1.

According to the multiple linear regression analysis we found, that the presence of MS was the only significant independent predictor of the RHI (beta=0.396, p<0.001).

Tab. 1. Characteristics of study populations.

	Multiple sclerosis	Controls	*p*-value
Number of subjects	46	31	
Age (years ±SD)	37.26±9.50	38.81±10.12	0.498
Male sex	8 (17.39%)	24 (77.42%)	≤0.001***
BMI (kg/m^2 ± SD)	22.83±4.51	28.20±2.93	≤0.001***
Number of smokers	5 (10.87%)	8 (25.81%)	0.086
RHI (median, IQR, range)	1.77, 0.71, 0.46–2.98	2.30, 0.98, 1.58–3.43	0.001***

(BMI: body mass index; RHI: reperfusion hyperemia index; SD: standard deviation; IQR: interquartile range).

DISCUSSION

In our study, values of RHI (assessed non-invasively using EndoPAT device), in the MS population were significantly lower than in healthy control group. This finding suggests worse endothelial function in MS group, although the control group seems to have greater burden of known vascular risk factors (significantly higher proportion of male sex, significantly higher BMI and non-significantly higher proportion of smokers). Nevertheless, we failed to find any significant correlation between measures of endothelial function and duration of the disease or disability (expressed as EDSS). We also failed to find any influence of current attack of the disease on endothelial function. We reported significantly higher RHI values, suggesting better endothelial function, in patients treated with glatirameracetate, than in patients treated with interferone beta-1b or in those without any DMT. Finally, the presence of MS seems to be the only significant independent predictor of the measures of endothelial function according to multiple linear regression model.

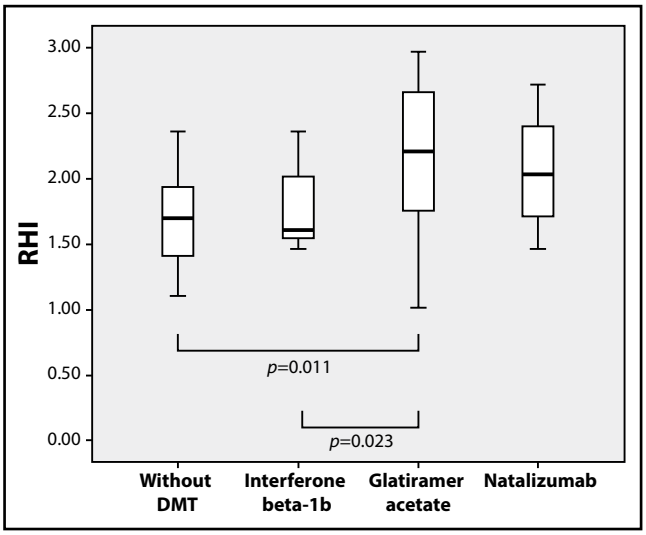

Fig. 1. Impact of disease modifying therapy (DMT) on reperfusion hyperemia index (RHI).

The causes of impaired endothelial function in MS patients may include several factors. First, endothelial dysfunction may be a consequence of multiple pathological mechanisms underlying SM. Chronic inflammatory response and oxidative stress, key-players in a process of atherogenesis, are also suspected to play a role in pathophysiology of MS (Blom *et al.* 2011; Maes *et al.* 2011; Sitia *et al.* 2010; Tenorio-Laranga *et al.* 2010). Second, spectrum of vascular risk factors in MS population differs from subjects without MS. Reduced physical activity, higher proportion of smokers were reported in previous studies. Also less typical vascular risk factors, like sleep disorders, can also play a role (Christiansen *et al.* 2010).

Third, treatment of MS patients with corticotherapy or DMT can participate in atherogenesis (Oberleithner *et al.* 2006).

Higher risk of vascular events in MS population was reported in several population-based studies. Patients with MS were reported to have about a 30% higher risk of cardiovascular mortality than the age-matched general population (Bronnum-Hansen *et al.* 2004; Koch-Henriksen *et al.* 1998). Another authors reported only 6% higher risk of dying of cardiovascular disease compared with healthy controls, but cerebrovascular disease was excluded from this analysis (Hirst *et al.* 2008). Allen *et al.* compared the prevalence of ischemic heart disease, myocardial infarction and ischemic stroke among 9,949 patients with MS and 19,898 patients without MS. MS patients were more likely to be hospitalized for ischemic stroke (OR 1.66, 95% CI 1.33–2.09) than the matched non-MS controls (Allen *et al.* 2008). Christiansen and co-workers conducted a large population-based cohort study involving 13,963 Danish citizens with MS and 66,407 age-matched and sex-matched controls. MS was associated with an increased risk of hospital admissions for ischemic stroke (IRR 1.92, 95% CI 1.27–2.71), myocardial infarction (1.84, 1.28–2.65), and heart failure (1.92, 1.27–2.90). These studies performed in New York and Denmark suggest, that patients with MS have an increased risk of ischemic stroke (Christiansen *et al.* 2010). The increased frequency of ischemic stroke and other ischemic cardiovascular events in MS might be caused by the endothelial dysfunction, which is believed to be the most important initial step in the process of atherogenesis.

The most widely accepted hypothesis for the pathogenesis of MS suggests autoimmune process leading to neuroinflammation and demyelination. Recently, new evidence showed that the immune system plays an important role in the pathogenesis of endothelial dysfunction. Endothelial dysfunction is an early step towards atherosclerosis and the immune system seems to be highly involved in atherogenesis as well as in MS (Sitia *et al.* 2010). Rheumatoid arthritis is another autoimmune disease in which increased cardiovascular morbidity and mortality have been noted (Khan 2010; Solomon 2010). Endothelial dysfunction has been described even in the very early stages of rheumatoid arthritis, probably as the result of inflammatory disease activity (Sodergren *et al.* 2010).

Several other factors can be involved in atherogenesis in MS patients. Alterations in endothelial function are mainly characterized by a reduction in the bioavailability of nitric oxide. Oxidative stress contributes to the development of endothelial dysfunction (Cai *et al.* 2000). Higher amounts of systemic and CNS oxidative stress have been reported in patients with MS than in healthy subjects (Choi *et al.* 2011; Koch *et al.* 2006; Tenorio-Laranga *et al.* 2010). Hyperhomocysteinaemia is another possible cause of endothelial dysfunction (He *et al.* 2010; Sekula *et al.* 2011). The cause of increase in homocysteine concentration is unknown,

but it occurs independently of vitamin B12, vitamin B6 or folate serum concentrations (Ramsaransing *et al.* 2006). Increased concentration of plasma homocysteine is described in MS patients (Russo *et al.* 2008; Sahin *et al.* 2007).

All of the traditional cardiovascular risk factors (dyslipidaemia, arterial hypertension, hyperglycemia and diabetes) are associated with endothelial dysfunction. Among the traditional vascular risk factors, smoking and reduced physical activity are most frequently reported in MS patients (Christiansen *et al.* 2010; Hernan *et al.* 2001). On the other hand, arterial hypertension, diabetes mellitus, dyslipidaemia and obesity generally do not seem to be associated with MS (Allen *et al.* 2008).

Our data suggested worse endothelial function in MS patients compared to healthy subjects, despite the following facts: the proportion of males in the control group of patients was significantly higher than in MS group (77.4% vs 17.39%) and the values of BMI were significantly higher in the group of healthy subjects than in MS group (22.83 ± 4.51 vs 28.20 ± 2.93). Both male sex and obesity are known vascular risk factors, so burden of known vascular risk factors seems to be higher in control group. Although, our population consisted of individuals with low burden of traditional risk factors, non-traditional risk factors like physical inactivity, spychosocial-stress or sleep disorders, that can also play role in atherogenesis, were not considered. Fatigue and depression are common symptoms, and neurological deficit in MS may lead to physical inactivity. Patients with MS also experience sleep disorders more frequently than control population (Brass *et al.* 2010).

Although the presence of MS seemed to be the only significant independent predictor of the measures of endothelial function in our study, we failed to reveal any relationship between the RHI values and disease characteristics, such as EDSS (r=–0.103, p=0.496) or disease duration (r=0.032, p=0.831). We assume, that inflammatory disease activity, involved in process of atherogenesis, does not have to be necessarily influenced with duration or clinical severity of the disease.

There are only limited data reporting the effect of DMT or corticosteroids on the endothelial function (Oberleithner *et al.* 2006). Although our data suggest better endothelial function in patients treated with glatirameracetate, than in patients treated with interferone beta-1b or in those without any DMT, low number of enrolled patients strongly limit interpretation of this fact. Patient with corticotherapy were excluded from our study, what could lead to a selection bias.

CONCLUSION

Results of our study suggest significant impairment of endothelial function in MS population compared to age matched control population with low burden of traditional vascular risk factors. Although the presence of MS seemed to be the only significant independent predictor of the measures of endothelial function in our study, also other aspects, like non-traditional risk factors or medication can participate in this process. Future longitudinal prospective randomized studies are needed to help to elucidate causes of impaired endothelial function in MS patients.

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Conflict if interest statement

The authors declare no conflict of interest.

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