LETTER TO THE EDITOR

ANTIBODIES TO THE 60 AND 65 KDA HEAT-SHOCK PROTEINS AND RISK OF PERIPHERAL ARTERY DISEASE

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Autoimmunity may be involved in atherogenesis. Beta2-glycoprotein I (beta2-gpI) and heat-shock proteins (Hsp) are found in the atherosclerotic plaque.[1,2] Our group has recently looked at the humoral response to these atheromatous components. Uniformly, IgA antibodies to beta2-gpI were associated with an increase in the risk of cerebral ischemia,[3] myocardial infarction,[4] and peripheral artery disease (PAD).[5]

In turn, IgG antibodies to the 60 kilodalton (kDa) human Hsp and to the 65 kDa *Mycobacterium bovis* Hsp behaved as risk factors for cerebral ischemia,[3] but not for myocardial infarction.[6] These intriguing findings led us to determine whether elevated levels of IgG anti-Hsp 60 and 65 are associated with an increased risk of PAD.

Seventy-seven patients with PAD and 93 controls were included in this case-control study. Diagnosis of PAD was based on clinical features (intermittent claudication, critical ischemia) and arteriographic changes (lesion, stenosis, occlusion). Controls with no symptoms of PAD were recruited from orthopedic wards. Risk factors evaluated in cases and controls included age, sex, race, hypertension, diabetes mellitus, smoking, and hypercholesterolemia. IgG antibodies to the human f Hsp 60 and to the *Mycobacteria bovis* Hsp 65 were detected by enzyme-linked immunoabsorbent assay (INOVA Diagnostics, Inc., San Diego, USA). Samples with optical densities above 0.5 were considered positive. Odds ratios (OR) were calculated by logistic regression.

The mean age was 61.5 years for cases and 47.5 for controls ($P < .001$). This age difference was adjusted by multivariate analysis. Caucasoids (strongly) and males (slightly) predominated in both groups. Among the known risk factors, hypertension yielded the strongest association with PAD (OR 12.1; 95% CI 5.8-30; $P < .001$). IgG anti-Hsp 60 antibodies were detected in 6.5% of cases but not in controls. This antibody was strongly associated with the risk of PAD (non-adjusted OR 14.2 after Agresti correction; 95% CI 1.5-infinite; $P = .02$). A negative association of IgG anti-Hsp 60 kDa with older age was obtained (OR 0.1; 95% CI 0-0.7; $P = .02$). IgG anti-Hsp 60 antibodies did not associate with any particular arteriographic pattern. The occurrence of IgG anti-Hsp 65 did not differ in cases and controls (adjusted OR 1.3; 95% CI 0.4-4.3; $P = .72$).

There have been few reports on anti-Hsp 60 or 65 and PAD. Wright et al. investigated the frequency of antibodies to Hsp of 60, 65, and 70 kDa in cases of PAD and renal vascular disease. Significant levels of anti-Hsp 60 antibodies were detected in patients with PAD, but not in those with renal disease. High levels of anti-Hsp 70 antibodies were found in both groups as compared to controls.[7]

This risk association of IgG anti-Hsp 60 kDa with PAD corroborates our previous findings in cerebral ischemia,[3] but differ from our results in myocardial infarction.[6] As opposed to our results on cerebral ischemia patients,[3] an association of IgG anti-Hsp 65 kDa was not confirmed in PAD patients.

In the atherogenic process, human Hsp 60, which is highly homologous to bacterial Hsp of 65 kDa, is synthesized by the endothelium as a response to cell injury.[8] The pathogenic role, if any, of anti-Hsp 60 or 65 in patients with cerebral ischemia, coronary disease, and PAD has yet to be clarified. Of interest, the antibody profile seems to differ in these various groups of atherosclerotic patients. A clinical use for these antibodies still requires confirmation. Epiphenomenon or not, antibodies to Hsp 60 or 65 might represent one of the links between autoimmunity and atheromatosis of the peripheral circulation.

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