Upregulation of the interleukin-8 system in hypercholesterolemic patients. Does inhibition of the mevalonate pathway lower interleukin-8 levels in the vessel wall?

Recently, Porreca et al. [1] reported a comprehensive study about peripheral blood mononuclear cell (PBMC) production of interleukin-8 (IL-8) and IL-8-dependent neutrophil function in hypercholesterolemic patients. The authors found an increase in the amount of IL-8 transcript in PBMC from hypercholesterolemic patients, in non-stimulated and in lipopolysaccharide (LPS) stimulated cultures, respectively. Correspondingly, a significant increase of IL-8 immunoactivity in the conditioned medium of PBMC from hypercholesterolemic subjects compared with controls was found. As the authors state, these results indicate an upregulation of the IL-8 system in dyslipidemic patients and provide further evidence for ongoing in vivo IL-8-dependent activation of inflammatory cells during hypercholesterolemia. Other groups reported that oxidized LDL and acetylated LDL were capable of inducing IL-8 synthesis in THP-1 macrophages, whereas native LDL had no impact on the production of the cytokine [2]. Furthermore, macrophages isolated from atherosclerotic plaques have been found to produce IL-8 [3]. In addition to its initial characterization as a proinflammatory cytokine, IL-8 has been shown to be synthesized by a variety of cell types and accordingly, to have a variety of functions. With regard to the constituents of the vessel wall, IL-8 plays an important role as a mitogen and chemoattractant for vascular smooth muscle cells (SMC) [4]. SMC migration can be stimulated by IL-8 to values 20-fold over those of controls. Since SMC proliferation and migration are crucial steps in the development of vessel wall thickening during atherosclerosis, these data raise the importance of IL-8 for the pathologic changes of the vessel wall.

As Porreca et al. [1] reported, increased levels of cholesterol are linked to an upregulation of the IL-8 system, which in turn may explain leukocyte activation during hypercholesterolemia and cause the inflammatory response during atherosclerosis. The authors also point out that inhibition of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase is an important means to inhibit atherosclerotic disease. In a rabbit atherosclerosis model, atorvastatin has been shown to reduce the neointimal inflammation, which could contribute to the stabilization of the atherosclerotic plaque [5]. However, it is widely unknown by which pathways HMG-CoA reductase inhibitors affect the pathogenesis of atherosclerosis apart from simply lowering the cholesterol. The interesting question now arises, whether the lowering of cholesterol levels is followed by a decrease of IL-8 levels. Monocytic THP-1 cells have been found to produce markedly decreased IL-8 levels following mitogenic stimulation when the cells were pretreated with HMG-CoA reductase inhibitors such as lovastatin or compactin at concentrations of 10 μmol/l [6]. In contrast, another group found that stimulated or non-stimulated IL-8 synthesis of PBMC is not affected by inhibition of the mevalonate pathway using simvastatin at doses up to 10 μmol/l [7]. However, IL-8 synthesis by blood cells does not directly reflect the IL-8 content in the vessel wall. Studies with Ca²⁺ channel blockers on SMC revealed increased levels of IL-8 due to the treatment with the compounds [8]. Among a variety of effects, Ca²⁺ channel blockers have been shown to downregulate the HMG-CoA reductase gene in fibroblasts [9]. In view of the unclear action of HMG-CoA reductase inhibitors on IL-8 expression, we studied the influence of lovastatin and simvastatin on IL-8 synthesis in human coronary SMC (hSMC) under in vitro conditions by applying 5–20 μmol/l of the compounds — doses comparable to those used in the above mentioned studies on blood cells — for an incubation time of 24–48 h. Intracellular IL-8 levels were determined by flow cytometry employing cell preparation methods as described previously [10] using a monoclonal antibody directed against recombinant human IL-8 (Endogen, Cambridge, MA). In contrast to the decreased production of IL-8 in human THP-1 monocytes under HMG-CoA reductase inhibitor treatment, we observed a marked increase of intracellular IL-8 levels up to 23-fold (relative fluorescence intensity) due to incubation with lovastatin or simvastatin (Fig. 1). Since HMG-CoA reductase inhibitors are known antiproliferative compounds, it may be discussed whether upregulation of IL-8 could be an autocrine production of a mitogen to overcome the cell...
Fig. 1. Flow cytometric analysis of intracellular IL-8 levels in hcSMC. The cells were cultured to subconfluency in the presence of DMEM media containing 10% FCS. Simvastatin (5 \text{mmol/l}) or lovastatin (5 \text{mmol/l}) were then added to the culture media and incubated for 48 h. Cells were fixed and permeated using 1% formaldehyde (5 min) following ice-cold pure methanol (30 min) and subsequent treatment with 0.1% Triton X-100 and 0.1% sodium citrate in phosphate buffered saline on ice (45 min), before IL-8 was indirectly stained using a monoclonal antibody specific for recombinant human IL-8. The data shown were generated in the same experiment.

References


18 October 1999

Jürgen R. Sindermann\textsuperscript{a,b}, Annette Schmidt\textsuperscript{a}, Günter Breithardt\textsuperscript{a,b}

\textsuperscript{a} Division of Molecular Cardiology, Institute for Arteriosclerosis Research, University of Münster, 48149 Münster, Germany

\textsuperscript{b} Department of Cardiology and Angiology, University of Münster Medical School, Domagkstraße 3, 48149 Münster, Germany

12 January 2000