PLASMA-DERIVED AND SYNTHETIC HIGH DENSITY LIPOPROTEINS MODULATE IL-6 PRODUCTION IN ENDOTHELIAL CELLS

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Several prospective studies have shown the existence of a strong inverse correlation between high density lipoprotein (HDL) plasma levels and the incidence of cardiovascular events (Wilson et al. 1988; Assmann and Schulte 1992; Gordon et al. 1989). The protective effect of HDL is believed to be due to their capacity to promote reverse cholesterol transport, the process by which cholesterol in peripheral tissues, including the arterial wall, is routed to the liver for the excretion from the body. Through this pathway HDL retard the formation of lipid-rich arterial lesions, thus preventing plaque rupture and coronary events (von Eckardstein et al. 2001). Several experimental evidences, however, suggest the existence of other HDL-mediated protective mechanisms that are independent from lipid metabolism. HDL are able *in vitro* to inhibit the expression of cellular adhesion molecules on the endothelium, monocytes transmigration, platelet aggregation and to improve fibrinolysis (Nofer et al. 2002). Interleukin-6 (IL-6) is a pleiotropic cytokine that acts on a wide variety of cell types. It has important regulatory functions in the immune system, it induces the production of the acute-phase proteins and it is involved in the regulation of differentiation, proliferation and survival of target cells. It has been demonstrated that patients suffering from angina, peripheral vascular diseases and congestive heart failure show higher IL-6 plasma levels if compared to healthy subjects (Woods et al. 2000). The aim of the present study was to evaluate if HDL are able to modulate IL-6 expression in HUVECs (human umbilical vein endothelial cells). HUVECs were incubated for 24 hours with increasing concentration of human plasma-derived HDL (0.25 – 2 mg/ml of protein content). After washing with PBS, lipopolysaccharide (LPS) was added at the concentration of 10 g/ml for 24 hours. Cell supernatants were collected at the end of the LPS incubation to evaluate IL-6 production. HDL were able to inhibit LPS-mediated IL-6 production in a dose-dependent way, with a maximum inhibition of 58.2% at the concentration of 2 mg/ml. To evaluate which component of HDL is responsible for this inhibitory effect, synthetic HDL were prepared (rHDL). rHDL were prepared from phosphatidylcholine and lipid free apolipoprotein A-I (apoA-I) or A-I_{Milano} (apoA-I_M); liposomes, made only by phosphatidylcholine (POPC), and lipid-free apoA-I were also prepared. When HUVECs were incubated with increasing concentration of rHDL (0.25 – 1 mg/ml of protein content), the TNF -induced IL-6 production was inhibited in a dose dependent way, comparable to what observed with plasma-derived HDL. The effect was independent from the kind of apolipoprotein used, since rHDL with apoA-I or apoA-I_M were equally effective. Neither lipid-free apolipoproteins nor POPC alone were able to inhibit TNF -induced IL-6 production, thus suggesting that an intact amphipatic -helix structured apolipoprotein is necessary for the inhibitory effect. The present study demonstrates that HDL are able to inhibit IL-6 production by endothelial cells *in vitro*. These results suggest a new possible mechanism for the protective effect of HDL against cardiovascular diseases, independent from their role in lipid metabolism.

References

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