Expression of Tob by Human Coronary Arteries and BMP-Mediated Stimulation of Coronary Artery Endothelial Cell Proliferation

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Background: Little is known regarding the role of bone morphogenetic protein (BMP) signaling that control vascular cell growth during normal homeostasis and in arterial pathologies, notably atherosclerosis and arterial calcification. Here we tested the hypothesis that Tob, an antiproliferative protein that negatively regulates BMP signaling, is expressed in human coronary arteries and in cultured human coronary artery endothelial cells (ECs) and that BMP2 stimulates proliferation in cultured ECs.

Methods and Results: Immunohistochemical localization showed abundant expression of Tob in ECs of normal human arteries. We therefore determined expression of Tob and BMP receptors (BMPR-IA, -IB, and -II) and Tob mRNA in ECs at baseline and stimulated with BMP2 (300 ng/mL for 6 hr). Furthermore, we performed the presence of the protesome inhibitor epoxomicin that enhances BMP signaling by preventing degradation of BMP signaling pathways. To test whether BMP2 stimulates proliferation, we determined whether Tob expression in BMP2 (+) and BMP2 (-) were affected by both BMP2 and epoxomicin. BMP2 may regulate EC proliferation, suggesting a possible role in the growth of ECs during normal homeostasis and in arterial pathologies. Thus, we investigated the role of Tob in EC proliferation and/or differentiation. To this end, we have investigated the role of Tob in EC proliferation via a mechanism that is not dependent on altered expression of Tob in response to BMP stimulation.

Conclusion: We have shown that homocysteine damaged endothelial cells even in clinically relevant concentrations through sustained exposure with increased Tob expression. And intracellular ROS production and NF-κB activation were observed and increased Tob expression might lead to inflammatory response in SAH-treated endothelial cells. These data identified novel mechanism of homocysteine inducing endothelial cell damage.

Changes in Innate and Adaptive Humoral Immune Responses and Indices of Atherosclerosis in Aging

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Background: Immune recognition against atherogenic cells is a promising therapy but the natural course of immune responses against oxLDL during aging is not known. We hypothesized that aging alters innate or adaptive immune responses to oxLDL modulating the progression of atherosclerotic plaque phenotype.

Methods: Western diet was sacrificed at 15-17, 36 weeks, and 50 weeks of age. Descending aorta was stenosed in mice for 20 months. Blood was collected for immunohistochemistry. Splenic cytokines were evaluated using RT-PCR.

Result: Aging was associated with increased atherosclerotic burden and collagen content with decreased macrophage and plaque lipid. MDA-LDL IgG increased in the 36 weeks group but reduced in mice with >52 weeks.

Conclusion: Immune response increase as antibody titers to Cox-LDL and PC is associated with increased plaque sizes and a more stable phenotype.

Atherogenic Low-Density Lipoprotein Impairs Vascular Endothelial Cell Survival by Disrupting the FGF2-PI3K-Akt Autoregulatory Loop

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Background: Atherogenic LDL, such as circulating electronegative LDL and oxidized LDL (oxLDL), can inhibit proliferation and induce apoptosis in vascular endothelial cells (EC). Fibroblast growth factor 2 (FGF2) stimulates phosphatidylinositol 3-kinase (PI3K), which in turn activates the Akt kinase, a protein kinase that regulates cell survival. We studied this design to investigated how oxLDL interferes with signal transduction along the FGF2-PI3K-Akt pathway.

Methods: The interrelationship between FGF2 and Akt was examined in cultured bovine aortic EC (BAEC). To investigate further the role of endogenous FGF2 in EC survival, BAEC[FGF2(+)] and BAEC[FGF2(−)] cell lines were established by stable transfection of BAEC with FGF2 sense and antisense cDNAs.

Results: In cultured BAEC, oxLDL (50 µg/ml) inhibited FGF2 transcription and Akt phosphorylation. Consistent with the cell-survival properties of APT, PI3K inhibitor wortmannin (25-200 nM) also inhibited FGF2 expression and induced apoptosis in a concentration-dependent manner. Stable overexpression of FGF2 in BAEC[FGF2(+)] greatly enhanced Akt phosphorylation, rendering the cells resistant to oxLDL-induced apoptosis. Expression of endogenous FGF2 in BAEC[FGF2(−)] failed to lead to expression of Tob, Akt autoregulatory loop

Conclusion: EC survival depends on continuous activation of the PI3K-Akt pathway by endogenous FGF2, which is required for its own induction in the manner of an autocrine.

Atherosclerotic Molecule-1 Expression

Jing Xia, Young-Lee Loo, Dae-Gyun Park, Hyo-Soo Kim, Youn-Bae Park, Seoul National University College of Medicine, Seoul, South Korea

Background & Aim: Atherogenic LDL, such as circulating electronegative LDL and oxidized LDL (oxLDL), can inhibit proliferation and induce apoptosis in vascular endothelial cells (EC). Fibroblast growth factor 2 (FGF2) stimulates phosphatidylinositol 3-kinase (PI3K), which in turn activates Akt, a protein kinase that regulates cell survival. We studied this design to investigate how oxLDL interferes with signal transduction along the FGF2-PI3K-Akt pathway.

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Thrombin and Histamine Stimulate Phosphorylation of Endothelial Nitric Oxide Synthase via an Akt-Independent, AMP-Activated Kinase-Dependent Pathway

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Background: The protein kinase Akt is involved in vascular development and several endothelial functions, including activation of endothelial nitric oxide synthase (eNOS) and promotion of endothelial cell survival. Recently we have found that Akt phosphorylation is inhibited by the G-protein activators thrombin and histamine these agonists stimulate phosphorylation of eNOS on Ser1179 The purpose of this study was to examine the role of other protein kinases in mediating this Akt-independent phosphorylation of eNOS.

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