The involvement of heparin-integrin interaction and nitric oxide signaling in the heparan sulfate synthesis by endothelial cells

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Endothelial cells are a source of physiologically important molecules synthesized and released to the blood and/or to the subendothelial extracellular matrix such as a heparan sulfate proteoglycan (HSPG) with antithrombotic properties. We have shown that heparin stimulates the synthesis of this HSPG in rabbit aorta endothelial cells. By confocal microscopy, heparin was detected bound on the extracellular matrix and was co-localized with fibronectin. Here some molecular mechanisms involved in the up-regulation of HSPG synthesis by heparin in endothelial cells were decoded. The cells were stimulated with heparin and the expression of HSPG and intracellular pathways were evaluated by a combination of methods involving confocal microscopy, flow cytometry, western blotting analyses, and [(35)S]-sulfate metabolically labeling of the cells. We observed that the up-regulation of HSPG synthesis evoked by heparin is dependent on the interaction of heparin with integrin since RGD peptide abolishes the effect. The activation of integrin leads to tyrosine-phosphorylation of focal adhesion-associated proteins such as FAK, Src. Moreover, heparin also induced eNOS (endothelial nitric oxide synthase) activation, as well as an increase in nitric oxide (NO) production. Finally, an eNOS inhibitor was able to abolish the effect in heparan sulfate synthesis. In conclusion, the heparin-induced up-regulation of HSPG expression is associated with the interaction with integrin, phosphorylation of focal adhesion proteins and NO pathways.

Word Keys: Endothelial cells, extracellular matrix, FAK, heparin, HSPG, nitric oxide.

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