Cell Biology of Arterial Proteoglycans

Thomas N. Wight

Although proteoglycans constitute a minor component of vascular tissue, these molecules have been shown to influence a number of arterial properties such as viscoelasticity, permeability, lipid metabolism, hemostasis, and thrombosis. A hallmark of early and late atherosclerosis is the accumulation of proteoglycans in the intimal lesions. Yet, it is not clear why this accumulation occurs. This article reviews the classes of proteoglycans synthesized by the two major cell types of the arterial wall—the endothelial and smooth muscle cell. Detailed consideration is then given to the modulation of proteoglycan metabolism and the role that proteoglycans play in a number of cellular events such as adhesion, migration, and proliferation—important processes in both the development and the pathogenesis of blood vessels. Last, the involvement of proteoglycans in two critical vascular wall processes—hemostasis and lipid metabolism—is reviewed, because these events pertain to atherogenesis.

This review emphasizes the importance of proteoglycans in regulating several key events in normal and pathophysiological processes in the vascular tissue. (Arteriosclerosis 9:1-20, January/February 1989)
Figure 1. A schematic model of the proposed structure of a typical proteoglycan monomer. The molecule consists of a central protein core to which are attached a variable number of glycosaminoglycans (GAG) side chains and various proportions of O-linked and N-linked oligosaccharides. Usually, one type of GAG chain is associated with a single protein core. The N-terminal end of the protein core may possess the capacity to interact with hyaluronic acid (HA-binding region), while other protein cores may contain hydrophobic regions facilitating their insertion into membranes. CS=chondroitin sulfate; DS=dermatan sulfate; HS=heparan sulfate; KS=keratan sulfate. Reproduced with permission from Singer et al. Recent advances in hematology 1985;4:1-24.

Figure 2. Electron micrographs of purified proteoglycan monomers prepared from different noncartilagenous sources. The chondroitin sulfate proteoglycan (CSPG) prepared from arterial smooth muscle cell cultures consists of a central core of ~275 nm to which are attached ~10 side chains averaging 70 nm in length. Another form of CSPG is present in the nerve terminal of electric organ (Carlson S, Wight TN. J Cell Biol 1987;105:3075-3086). This molecule possesses a central core of ~345 nm with 20 to 25 side projections averaging 113 nm in length. Smaller proteoglycans containing dermatan sulfate (DSPG) also are synthesized by arterial smooth muscle cells. These molecules possess a central core of ~100 nm and only one or two side projections of varying length (40 to 70 nm). Proteoglycans containing heparan sulfate (HSPG) vary enormously in size. The HSPG presented in this figure has been isolated from the plasma membrane of epithelial cells (Rapraeger et al. In: Wight TN, Mecham RP, eds. Biology of proteoglycans. New York: Academic Press, 1987:129-154). It consists of a central core of ~130 nm with 3 to 4 side chains of varying length (50 to 70 nm), x83 000. Bar=0.1 μm.

Vascular Cell Proteoglycans

Endothelial Cells

Vascular endothelial cells synthesize and secrete both heparan sulfate and dermatan sulfate containing proteoglycans. However, it is still not clear why proteoglycans accumulate and what specific effects these molecules have on events associated with the pathogenesis of these diseases. This review focuses on these two questions with emphasis on recent developments in the field of arterial proteoglycans, since studies prior to 1980 have already been reviewed.3
Figure 3. Light micrograph illustrating that the narrow intima of a normal blood vessel stains more intensely with (A) alcian blue and (B) a monoclonal antibody against aortic CSPG than the underlying medial layer. Vessels undergoing intimal hyperplasia (early atherosclerosis) possess thickened intimas that also stain intensely with alcian blue (C) and a monoclonal antibody against aortic CSPG (D). ×281. Reproduced with permission from Wight et al. In: Wight TN, Mecham RP, eds. Biology of proteoglycans. Orlando, FL: Academic Press, 1987:267-300. Bar=50 μm.

glycans. These molecules differ in size and in ability to interact with other molecules. For example, there are a number of size classes of HSPG present in endothelial cultures. One form is hydrodynamically large (~400 kd) and sediments in cesium chloride at low buoyant density. It appears that the larger species of HSPG exists as disulfide bonded aggregates, linked either to itself or to other matrix components, as has been demonstrated for HSPG in a mouse tumor basement membrane. In addition, a HSPG (~250 to 400 kd) from cultured vascular endothelial cells that exhibits hydrophobic properties interacts specifically with antithrombin III and, therefore, is thought to play a critical role in providing a nonthrombogenic surface for the vascular endothelium (see also Hemostasis section).

Structural differences among the subclasses of endothelial derived HSPGs may arise in part from the synthesis of unique HSPGs on cores that are distinct products of separate genes. For example, Stow and coworkers have shown that antibodies raised against a putative hepatocyte plasma membrane HSPG failed to stain glomerular basement membrane, staining only the surfaces of kidney epithelia and endothelia, while the reverse was true for an antibody raised against HSPG from basement membrane. The most recent study by this group has shown that certain cell types in culture synthesize two forms of HSPG, one that possesses hydrophobic properties consistent with a membrane location and another that is nonhydrophobic and reacts only with the antibasement membrane HSPG antibody. Others have shown similarly that structurally distinct cell surface and basement membrane associated HSPGs found in mouse mammary epithelial cells also are immunologically distinct.

A recent study of the high- and low-density forms of HSPG in the Engebreth-Holm-Swarm (EHS) basement membrane producing tumor indicates that the two forms contain distinctly different core proteins and glycosaminoglycan chains. Alternatively, posttranslational or postsecretional processing also may generate some of the multiple HSPG species, either as a mechanism for targeting HSPGs with different functions to different cellular and extracellular compartments or as a result of
HSPG turnover. For example, Ledbetter et al. identified a 400 kd precursor protein that was converted to a large, low-density HSPG in the EHS tumor system. Similarly, Klein et al. recently presented evidence that the glomerular basement membrane HSPG was derived from a larger HSPG precursor. Other studies following the secretion, uptake, and intracellular degradation of single HSPG species indicate that several species of HSPG are generated by turnover mechanisms operating in cell culture. Thus, it appears that endothelial cells are the source of a variety of HSPG subclasses. Their origin and specific function(s) await further study.

Vascular endothelial cells also synthesize a family of proteoglycans that contain dermatan sulfate. These proteoglycans contain glycosaminoglycan chains of low iduronic acid content (10%) when the cells are cultured on plastic, but a higher percentage of iduronic acid when cultured on collagen gels. At least three separate subclasses have been identified. One species appears to be a long-lived (t1/2 = 40 hours) high molecular weight complex consisting of DSPG covalently linked to fibronectin via transglutaminase cross-linking. Two other DSPGs can be distinguished in these cultures (M, ~ 220 000 and ~ 130 000); these resemble similar subclasses that have been designated as DSPG I and II in cartilage and a variety of other tissues (reviewed in references 56 and 57).

It is now clear that a significant proportion of the proteoglycans synthesized by endothelial cells are deposited as basement membranes. For example, an antibody raised against the large low density HSPG in the EHS tumor immunostains the matrix synthesized by cultured bovine aortic endothelial cells and vascular basement membranes. The role of proteoglycans as part of this specialized structure appears to be at least twofold. One role is to serve as a structural organizer of the other components of the basement membrane (such as type IV collagen, laminin, entactin, and fibronectin) by virtue of the ability of proteoglycans to interact at specific sites within each of these molecules. Another role is to contribute to the selective filtration barrier function of basement membrane by conferring a network of fixed charges to this structure. Alterations in the proteoglycan content of basement membranes severely compromise the function of this filtration barrier. Basement membranes in diabetic tissues have a reduced HSPG content and are undersulfated. Such alterations are thought to contribute to the increased vascular wall permeability frequently seen in diabetes.

**Smooth Muscle Cells**

Unlike endothelial cells that synthesize predominantly HSPGs, vascular smooth muscle cells synthesize and secrete principally CSPGs and DSPGs with considerably less HSPG (Figures 6 and 7). In general, these proteoglycans resemble the bulk of the proteoglycans present in intact arteries. Chondroitin sulfate proteoglycan is a major arterial proteoglycan. Biochemical and ultrastructural studies of CSPG synthesized by cultured monkey arterial smooth muscle cells indicate that they are hydrodynamically large molecules (M, 2 x 10^6) that consist of a core glycoprotein measuring 220 nm to 320 nm in length (M, ~ 400 000 to 600 000). Other studies following the secretion, uptake, and intracellular degradation of single HSPG species indicate that several species of HSPG are generated by turnover mechanisms operating in cell culture. Thus, it appears that endothelial cells are the source of a variety of HSPG subclasses. Their origin and specific function(s) await further study.
aggregate formed in blood vessels is stabilized by at least one accessory link protein.

Immunocytochemical studies reveal that a large CSPG is restricted to the interstitial matrix of arterial smooth muscle cell cultures and blood vessels and is not part of other matrix components such as collagen, elastic fibers, and basal laminae (Figure 3). However, a smaller CSPG that has hydrophobic properties and is immunologically distinct from the large interstitial CSPG is present in nonhuman primate arterial smooth muscle cell cultures and may be associated with the plasma membrane of these cells. A CSPG has been identified on the surface of rat arterial smooth muscle cells, as well as associated with membranes of other cell types. At present, it is not clear what role these membrane-associated proteoglycans play in the physiology of vascular cells, but they most probably are involved in mechanisms associated with cell adhesion, migration, and proliferation (see next section).

A second population of smaller proteoglycans containing dermatan sulfate also is synthesized by arterial smooth muscle cells. Electron microscopy of purified DSPG from these cells reveals a short core glycoprotein (100 nm) with 1 to 2 glycosaminoglycan side chains of $M_r \sim 40,000$ to 60,000 (Figure 2). The core glycoprotein of this class is $\sim 45,000$ (Figure 7). These cells appear to synthesize two types of DSPG. One species is present in the culture medium and contains a low percentage of iduronic acid ($<20\%$). The other class is deposited in the cell layer matrix and contains a much higher percentage of iduronic acid ($\sim 50\%$); it is the predominant proteoglycan deposited when arterial smooth muscle cells are cultured on collagen gels. Two types of DSPG that vary in iduronic acid content also have been found in intact blood ves-
Figure 6. Gel filtration chromatography of $^{35}$SO$_4$-radiolabeled arterial smooth muscle cell layer extracts at quiescence (A), during growth stimulation (B), or grown in the presence of collagen gels (C). In A, peak I contains a mixture of two hydrophobic proteoglycans, one that contains heparan sulfate chains and one that contains chondroitin sulfate (CS) chains. Peak II contains chondroitin sulfate proteoglycan and Peaks III and IV contain dermatan sulfate proteoglycans. Inset in B represents total incorporation of $^{35}$SO$_4$ into proteoglycan expressed on a per cell basis, at quiescence (open bar) or growth stimulation (shaded bar). Inset in C represents similar radioactivity from cells on plastic (open bar) or within collagen gels (shaded bars).

Unlike CSPG, the DSPGs are closely associated with the periphery of collagen fibrils at regular intervals in blood vessels. Considerable biochemical and morphological evidence indicates that DSPG is capable of binding to collagen fibrils and influencing collagen fibrillogenesis in some tissues. A recent study demonstrated that collagen fibril diameter increases from vessel intima to adventitia and is correlated with decreasing proteoglycan concentration. Tissues enriched in collagen, such as advanced atherosclerotic plaques, also are enriched in DSPG (Figure 4).

Dermatan sulfate proteoglycans synthesized by arterial smooth muscle cells resemble similar molecules found in a number of tissues including sclera, tendon, skin, cartilage, and blood vessels. In vitro translation experiments with RNA from arterial smooth muscle cells and an antibody against the small DSPG present in cartilage and skin indicates that the core protein of the arterial smooth muscle cell DSPG is ~41 kd and is immunologically related to both skin and cartilage DSPG. In addition, recent experiments indicate similar core protein sizes for DSPGs that are synthesized by arterial smooth muscle cells and skin fibroblasts. Interestingly, however, the DSPG synthesized by arterial smooth muscle cells is larger, due to longer glycosaminoglycan chains, and contains less iduronic acid than the DSPG secreted by skin fibroblasts.

Both the large (CSPG) and the small (DSPG) proteoglycans synthesized by arterial smooth muscle cells in culture appear to be secreted at similar rates with a $T_{1/2}$ of ~8 minutes (i.e., transit time from Golgi to the cell surface).
The capacity of endothelial cells to adhere to their connective tissue substrata is critical to the maintenance of normal vascular hemostasis. In fact, temporary detachment of endothelial cells has been postulated as one of the early events in the genesis of atherosclerotic lesions. Experimental removal of endothelial cells leads to marked atherosclerotic development in experimental animals. Thus, it is important to understand those factors involved in endothelial cell adhesion. Proteoglycans have been implicated in the adhesion of a variety of cells to their substratum. Most cells, including vascular endothelial cells, interact with their substrata in specialized areas of the cell known as focal adhesion sites. These sites are enriched on the ECM side in attachment proteins, such as fibronectin and heparan sulfate proteoglycans, and on the cytoplasmic side in actin microfilaments. Colocalization of HSPG with cytoskeletal elements such as actin suggests that proteoglycans may influence cell adhesion through association with the cytoskeleton. Recent studies have shown that the integrity of actin stress filaments is critical for endothelial cell adherence when exposed to shear stress. Although it is not completely certain as to how cell-associated HSPG mediates cellular adhesion, one theory is that proteoglycans may stabilize the adhesion process by interacting with specific proteoglycan binding domains in the attachment protein, such as fibronectin, thus stabilizing the interaction of fibronectin with its cell surface receptor. It is of interest that removal of HSPG from endothelial cells does not detach cells but does prevent cells from reattaching. Such studies indicate that endothelial cells, like many other cell types, possess multiple adhesion mechanisms. The fact that heparin and heparan sulfate treatment of deposited endothelial-derived extracellular matrix significantly inhibits endothelial cell adhesion supports the hypothesis that proteoglycans may be facilitating attachment by interacting with other adhesive molecules. Late passage endothelial cells that detach more readily than early passage endothelial cells also are characterized by less proteoglycan at their attachment sites.

While HSPG appears to promote cell attachment for some cells, other proteoglycans have opposite effects. For example, both CSPG and DSPG inhibit the adhesion of a variety of cells to either fibronectin or collagen. These proteoglycans may destabilize focal cellular adhesions, possibly interfering with specific cell binding sites in the adhesive glycoproteins. For example, the proteoglycan may bind to the adhesion protein and sterically interfere with the binding of the protein to its cell surface receptor. Alternatively, the binding of proteoglycan may change the configuration of the protein, causing a decrease in its association constant for the receptor. Evidence for a conformational change upon proteoglycan binding has recently been reported for plasma fibronectin. Thus, it appears that proteoglycans influence adhesion by modulating the activities of the primary molecular components involved in this process.

The capacity of endothelial cells to adhere to their connective tissue substrata is critical to the maintenance of normal vascular hemostasis. In fact, temporary detachment of endothelial cells has been postulated as one of the early events in the genesis of atherosclerotic lesions. Experimental removal of endothelial cells leads to marked atherosclerotic development in experimental animals. Thus, it is important to understand those factors involved in endothelial cell adhesion. Proteoglycans have been implicated in the adhesion of a variety of cells to their substratum. Most cells, including vascular endothelial cells, interact with their substrata in specialized areas of the cell known as focal adhesion sites. These sites are enriched on the ECM side in attachment proteins, such as fibronectin and heparan sulfate proteoglycans, and on the cytoplasmic side in actin microfilaments. Colocalization of HSPG with cytoskeletal elements such as actin suggests that proteoglycans may influence cell adhesion through association with the cytoskeleton. Recent studies have shown that the integrity of actin stress filaments is critical for endothelial cell adherence when exposed to shear stress. Although it is not completely certain as to how cell-associated HSPG mediates cellular adhesion, one theory is that proteoglycans may stabilize the adhesion process by interacting with specific proteoglycan binding domains in the attachment protein, such as fibronectin, thus stabilizing the interaction of fibronectin with its cell surface receptor. It is of interest that removal of HSPG from endothelial cells does not detach cells but does prevent cells from reattaching. Such studies indicate that endothelial cells, like many other cell types, possess multiple adhesion mechanisms. The fact that heparin and heparan sulfate treatment of deposited endothelial-derived extracellular matrix significantly inhibits endothelial cell adhesion supports the hypothesis that proteoglycans may be facilitating attachment by interacting with other adhesive molecules. Late passage endothelial cells that detach more readily than early passage endothelial cells also are characterized by less proteoglycan at their attachment sites.

While HSPG appears to promote cell attachment for some cells, other proteoglycans have opposite effects. For example, both CSPG and DSPG inhibit the adhesion of a variety of cells to either fibronectin or type I collagen. These proteoglycans may destabilize focal cellular adhesions, possibly interfering with specific cell binding sites in the adhesive glycoproteins. For example, the proteoglycan may bind to the adhesion protein and sterically interfere with the binding of the protein to its cell surface receptor. Alternatively, the binding of proteoglycan may change the configuration of the protein, causing a decrease in its association constant for the receptor. Evidence for a conformational change upon proteoglycan binding has recently been reported for plasma fibronectin. Thus, it appears that proteoglycans influence adhesion by modulating the activities of the primary molecular components involved in this process.

Table 1. Arterial Smooth Muscle Cell

<table>
<thead>
<tr>
<th>Type</th>
<th>Size* (Kav)</th>
<th>Characteristic</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSPG (1)</td>
<td>0.31</td>
<td>aggregating</td>
<td>interstitial matrix</td>
</tr>
<tr>
<td>CSPG (2)</td>
<td>0.60</td>
<td>hydrophobic</td>
<td>plasma membrane (?)</td>
</tr>
<tr>
<td>DSPG (IdA)</td>
<td>0.65</td>
<td>iduronic acid-rich</td>
<td>collagen</td>
</tr>
<tr>
<td>DSPG (GIA)</td>
<td>0.65</td>
<td>glucuronic acid-rich</td>
<td>?</td>
</tr>
<tr>
<td>HSPG (1)</td>
<td>0.60</td>
<td>hydrophobic</td>
<td>plasma membrane</td>
</tr>
<tr>
<td>HSPG (2)</td>
<td>0.60</td>
<td>detergent-resistant</td>
<td>matrix, basement membrane (?)</td>
</tr>
</tbody>
</table>

*Hydrodynamic size (elution position) on a Sepharose CL-2B column (4M GuHCl)

CSPG = chondroitin sulfate proteoglycan, DSPG = dermatan sulfate proteoglycan, HSPG = heparan sulfate proteoglycan.

(Yeo and Wight, unpublished observations), which is comparable to the secretion kinetics of other proteoglycan secreting cells such as rat ovarian granulosa cells, chondrocytes, and skin fibroblasts. However, it is of interest that these two types of proteoglycans are internalized by arterial smooth muscle cells at different rates, with DSPG exhibiting rapid, high-affinity, and saturable kinetics of uptake, whereas CSPG is taken up more slowly and by a low-affinity endocytic process. These data indicate that the turnover of different families of proteoglycans proceed at different rates.

A third class of proteoglycan synthesized by arterial smooth muscle cells contains heparan sulfate chains. This class represents a minor fraction of the total proteoglycans synthesized by these cells and is usually found in larger amounts in the cell layer matrix (20%) than in the medium (<5%). The majority of HSPG present in the cell layer elutes as a complex in the void volume of a Sepharose CL-2B column but shifts to an included position of Kav 0.6 when extracted in the presence of detergent. The HS chains are long (Mr ~ 40 000 to 60 000) and both N- and O-linked oligosaccharides are attached to the protein core. This molecule has an affinity for hydrophobic binding resins and may represent an in vivo state.
chains. This property may enable membrane forms of HSPG to self-associate and thus have a stabilizing influence on cell-cell association. Growing cells, which should be less adhesive, contain cell-surface HSPG with heparan sulfate chains that show little tendency to self-associate. Proteoglycans also appear to induce the formation of gap junctions in primary liver cultures by stimulating the synthesis of a specific gap junction protein. It remains to be seen whether a similar situation exists for other types of cells such as vascular endothelial and smooth muscle cells.

Cell Migration

The migration of vascular endothelial cells in response to injury or during angiogenesis and of smooth muscle cells during intimal hyperplasia are critical events in vascular wall development and disease. The migration of vascular cells is thought to be highly dependent on their ability to synthesize and secrete certain components of the extracellular matrix. For example, Madri and Stenn have shown that changes occur in the distribution of collagens and laminin during vascular endothelial cell sprouting and migration in vitro and have suggested that continued secretion of collagen is required for effective migration of these cells. Kinsella and Wight have demonstrated that endothelial cells induced to migrate by wounding increase their synthesis of proteoglycan by fourfold and shift from synthesizing a primarily HSPG-rich endothelial matrix to synthesizing a CSPG/DSPG-rich matrix (Figure 8). Autoradiographic evidence revealed that incorporation of proteoglycan precursor (35S-sulfate) was greatest in migrating cells near the wound edge. These results support the concept that proteoglycans participate in the migratory response of cells. A number of recent studies indicate that the migration of neural crest cells is inhibited by the addition of CSPG when added to collagen or fibronectin matrices, suggesting decreased cell-substratum adhesion. It is tempting to speculate that the glucuronic acid-rich DSPG synthesized by endothelial cells after wounding is involved in the facilitation of migration by interfering with the cell's adhesive mechanism. Oohira et al. noticed the loss of HSPG in cultures of sprouting (migrating) endothelial cells and Ausprunk et al. presented histochemical evidence suggesting that HSPG is depleted at the tips of growing capillaries. Other factors known to influence angiogenesis, such as interleukins, also exhibit a marked effect on proteoglycan deposition by vascular endothelial cells.

Smooth muscle cell migration from the media to the intima of blood vessels is an important component of the early vascular response to injury and, as such, may play a pivotal role in atherogenesis. Little is known about the factors that control this process. Recent studies by Majack and Clowes have shown that heparin inhibits arterial smooth muscle cell migration in vitro in a dose-dependent and reversible fashion, but hyaluronate and chondroitin or dermatan sulfate do not. Furthermore, this effect was specific for arterial smooth muscle cells, since similar treatments had no effect on cultured bovine aortic endothelial cells or Swiss 3T3 cells. In fact, heparin stimulates the migration of bovine capillary endothelial cells. The mechanism for the antimigratory effect of heparin on smooth muscle cells is not yet known.

Such studies indicate that vascular cell migration is accompanied by qualitative and quantitative changes in proteoglycans. It remains to be shown whether such changes regulate this cellular response and if so, how these changes affect the "migratory machinery" of the cell.

Cell Proliferation

Two key events in the development of the atherosclerotic plaque are the proliferation of arterial smooth muscle cells and the deposition of components of the extracellular matrix. An important question is whether these two events are related. Recent studies indicate that quiescent arterial smooth muscle cells increase their synthesis of proteoglycan when stimulated to divide (Figure 6B) and this increase occurs principally during the G1 phase of the cell cycle. The activity of several enzymes involved in the synthesis of chondroitin sulfate chains,
such as xylosyltransferase, N-acetylgalactosaminyl transferase I, and two sulfotransferases, increase during the proliferative phase in arterial smooth muscle cell cultures. These results suggest that stimulation may involve, in part, an increased capacity of the cells to synthesize the glycosaminoglycan moiety of the proteoglycans. This increase in synthesis is not restricted to proteoglycans, since other matrix molecules such as collagen and thrombospondin also increase during growth stimulation. These changes may, in part, account for the initial accumulation of matrix molecules during the proliferative phase of atherosclerosis. At present, it is not clear whether these synthetic modulations are necessary for proliferation or are a consequence of the proliferative response. It is of interest that growth stimulation of density-arrested Balb 3T3 cells is accompanied by an increase in the capacity of the cells to glycosylate proteins, and interference with this capacity markedly inhibits some cells from progressing through their cell cycle. Such results suggest that cell cycle progression may be regulated in part by the expression of specific glycoproteins and/or proteoglycans.

Proteoglycans have also been implicated in the regulation of cell proliferation. For example, HSPG derived from postconfluent endothelial and smooth muscle cells inhibits the proliferation of arterial smooth muscle cells, and this inhibitory activity is highly dependent on the presence of a platelet heparitinase in serum. This enzyme generates dodecasaccharide heparan sulfate fragments, which are active as growth inhibitors. Similar heparitinase activity is also present in resident cells of the arterial wall, including smooth muscle cells as well as blood-derived cells such as lymphocytes, macrophages, and neutrophils. In fact, extravasation of blood cells into the arterial wall is correlated with level of heparitinase activity. The fact that lymphocytes, macrophages, and neutrophils are present in lesions of atherosclerosis indicates a potential for the generation of proliferation-inhibitory proteoglycan fragments in vascular disease. Smooth muscle cells possess specific high-affinity receptors for heparin/heparan sulfate and can internalize this bound glycosaminoglycan. These results raise the interesting possibility that heparin/heparan sulfate and heparin/heparan sulfate fragments may suppress the growth of smooth muscle cells at specific intracellular sites. Additional studies support this possibility. For example, HSPG is present on the hepatocyte cell surface as a phosphatidylinositol-linked proteoglycan. This HSPG is metabolically processed on the cell surface with the loss of the diacylglycerol portion of the phosphatidylinositol and is internalized by receptor-mediated endocytosis. A part of the HSPG is then processed in a nonlysosomal compartment of the cell, and selected fragments of heparan sulfate chains containing an unusual sulfated glucuronic acid appear in the nucleus without the core protein. Changes in the structures and amounts of the nuclear heparan sulfate are accompanied by changes in the growth behavior of hepatocytes. Such studies indicate that the internalization of HSPG by a cell may, in part, regulate the growth of that cell.

Thus, evidence continues to accumulate that some forms of HSPG act as growth modulators, yet the precise mechanism(s) is still not known. A number of studies have shown that heparin alters the secretory phenotype of cultured arterial smooth muscle cells. For example, heparin induces the synthesis of a 60,000 molecular weight collagen-like protein at 18 to 24 hours after the addition of the heparin molecule. Other proteins of lower molecular weight (i.e., 35, 37, 39, kd), believed to be apolipoprotein (apo) E containing proteins, are induced within 4 to 8 hours of heparin exposure. No direct correlation between heparin regulation of these proteins and growth inhibition has been established, but it is noteworthy that these synthetic changes occur at dosages that are normally growth-inhibitory.

A recent finding that heparin decreases thrombospondin concentration in the extracellular matrix of cultured arterial smooth muscle cells is of interest because antibodies to thrombospondin were found to inhibit DNA synthesis in these cells. In addition, thrombospondin and the mitogen epidermal growth factor (EGF) act synergistically to stimulate DNA synthesis by smooth muscle cells, and this effect is inhibited by heparin. Thus, it may be that certain components of the extracellular matrix are necessary for cellular proliferation and that other matrix components, such as proteoglycans, influence the availability of such molecules. The fact that cell-associated HSPG can serve as a receptor for thrombospondin supports this possibility. It is also possible that heparin-like molecules regulate growth by interfering with the response to specific mitogens. For example, heparin reduces the number of EGF receptors on smooth muscle cells. Thus, the effect of heparin on growth may be due to a combination of effects on the matrix surrounding the cells and on the mitogenic machinery of the cell itself. Heparin also is effective in reducing smooth muscle cell proliferation in vivo after arterial injury and is thus capable of counteracting the effects of local and blood borne growth factors. Associated with this cessation of growth is an increase in the concentration of proteoglycans in injured arteries. In vitro studies have also shown that arterial smooth muscle cell cultures accumulate more proteoglycan in the presence of heparin. The nature of this modulation is currently under study. The reader is also referred to a recent review that discusses heparin's effect on other arterial processes.

Although most of the evidence indicates that the ability of heparin and heparan sulfate to inhibit growth is not caused by their interaction with peptide growth factors to inactivate them, a variety of peptide growth factors with mitogenic activity toward smooth muscle cells, fibroblasts, and endothelial cells display heparin binding affinity. Endothelial cell growth factor (ECGF) is a polypeptide mitogen that has been purified and characterized from bovine neural tissue. This mitogen supports the proliferation and serial propagation of human endothelial cells in vitro. Heparin binds to ECGF and potentiates the mitogenic activity of this polypeptide. The effectiveness of the interaction with heparin correlates with the ability of the mitogen to stimulate endothelial cell proliferation. Heparin also releases ECGF from the surface of cultured endothelial cells. Although the mechanism of heparin action is unclear, the synergistic activity suggests struc-
tural interactions between the carbohydrate and mitogen. It is possible that heparin potentiates the activity of EGF by causing the mitogen to assume a conformation with increased affinity for its receptor. Likewise, it is possible that the receptor requires a heparin-ECGF complex for an "ideal fit." Heparin also exhibits affinity for other growth factors such as platelet-derived growth factor, insulin-like growth factor, and fibroblast growth factor. Such interactions may effectively compartmentalize and retain growth factors for maximal activity. For example, Vlodavsky and colleagues have shown that heparin-binding FGF-like growth factors, which are angiogenic, are sequestered in the extracellular matrix by virtue of their binding to HSPG. Destruction of the heparan sulfate chains by heparinase efficiently releases the growth factor from the extracellular matrix of basement membranes. Similar sequestration of growth factors by proteoglycan has also been observed in bone marrow.

Collectively, these studies indicate that the glycosaminoglycan heparin, which is derived from mast cells, is capable of potentiating the action of growth factors in vitro. The presence of mast cells at the leading edge of developing blood vessels and the fact that heparin stimulates the growth of endothelial cells in vitro and angiogenesis in vivo suggests that mast cell heparin is important in modulating vascular cell behavior in vivo. Consistent with this suggestion is the observation that mast cell granules containing heparin can be internalized and degraded by endothelial cells and can cause proliferation of microvascular endothelial cells. It also may be that heparin-like molecules in the form of HSPG synthesized by endothelial or smooth muscle cells are effective in binding and potentiating the action of these different mitogens as suggested by the studies cited above. As discussed earlier, the HS chains in HSPG have some structural features in common with heparin derived from mast cells. In fact, a recent study demonstrates heparin sequences in the heparan sulfate chains of an endothelial cell proteoglycan. Anticoagulant HS has been isolated from vascular tissue that was shown to be free of mast cells. However, Imamura and Mitsui recently demonstrated that heparan sulfate and heparin exhibited opposing activities on the growth of human umbilical vein endothelial cells in the presence of acidic and basic FGF. Such results illustrate that the two molecules may differ in their activities associated with growth.

Proteoglycans also may act as receptors for growth regulatory substances. For example, Fransson et al. have shown that the core protein of a cell surface HSPG in fibroblasts binds transferrin and appears to have a structure nearly identical to the transferrin receptor. In addition, thrombospondin binds to the surface of vascular endothelial cells in a receptor-like fashion and this binding is inhibited by heparin, suggesting that thrombospondin associates with the surface of endothelial cells in a HSPG-dependent manner. The importance of thrombospondin in regulating smooth muscle cell growth suggests a growth regulatory role for the HSPG present on the surface of smooth muscle cells. It remains to be determined whether thrombospondin associates with the surface of smooth muscle cells as it does with endothelial cells. Majack et al. made the interesting suggestion that surface thrombospondin, presumably bound to a membrane form of HSPG, interact with certain proteases to facilitate the matrix degradation and remodeling that accompanies cell proliferation. Heparin, which inhibits proliferation, also releases thrombospondin from the cell surface. Such loss of thrombospondin may result in dissociation of the protease from the cell surface and loss of the capacity of matrix degradation to permit cell division. Such speculation awaits proof.

Recent studies have shown that the high molecular weight receptor for transforming growth factor beta (TGF/β) is a proteoglycan containing heparan sulfate and CS chains. These studies illustrate that cell surface-associated proteoglycans may play critical roles in the regulation of cell growth and differentiation.

Modulation of Proteoglycan Metabolism

A number of other factors are capable of modulating proteoglycan metabolism by vascular cells in the absence of cell proliferation and migration. For example, TGF/β, a platelet product, stimulates CSPG synthesis by human arterial smooth muscle cells without the stimulation of cell proliferation. This effect is cell-specific in that this platelet product had no effect on proteoglycan synthesis by vascular endothelial cells. The type of extracellular matrix surrounding vascular cells also influences the metabolism of proteoglycans. For example, when arterial smooth muscle cells are cultured on hydrated collagen gels (type I collagen), they decrease their overall proteoglycan accumulation (expressed on a per cell basis) but increase the amount of iduronic acid-rich dermatan sulfate present in the extracellular matrix (Figure 6C). Pulse-chase studies indicate that the accumulation of this specific proteoglycan within the cell layer is due partly to decreased turnover and partly to increased synthesis. Other extracellular factors shown to influence proteoglycan synthesis by cultured vascular cells include oxygen, prostaglandins, and lipids. These studies indicate that a variety of factors are capable of influencing proteoglycan metabolism by vascular cells.

Hemostasis

Endothelial cells provide a nonthrombogenic lining for the vascular system by secreting or sequestering factors involved in maintaining blood fluidity such as plasminogen activators, prostacyclin, and thrombomodulin. In addition, endothelial cells synthesize and secrete heparin-like HSPGs that bind to antithrombin III and enhance the inactivation of thrombin, inhibiting the clotting cascade. Heparin and heparan sulfate are related molecules that are part of a large family of heterogeneous proteoglycans. Heparin is distinguished from heparan sulfate in that it is more highly charged, appears to occur entirely in mast cells, and is an effective anticoagulant. However, heparin has many structural features in com-
mon with heparan sulfates. Both mast cell heparins and HSPGs contain anticoagulant and nonanticoagulant species. Generally, heparin contains more N- and O-sulfate and more iduronic acid than do HSPGs. Heparan sulfates, on the other hand, have a more ubiquitous animal and tissue distribution. Refer to Hovingh et al. for a discussion of the relationship between these two sets of molecules.

The binding of heparin and heparan sulfate to antithrombin III occurs at lysyl residues in the antithrombin molecule and is thought to involve a specific molecular domain composed of the tetrasaccharide sequence: IdA (GlcA)—GlcNAc—6-O-SCV-GlcA—GlcN—SO3-3-O-SO3, which is absent in nonanticoagulant heparin or heparan sulfate (for reviews, see references 38 and 205). This binding is responsible for a 1000-fold acceleration of enzyme-inhibitor complex formation. It is of interest that other purified hemostatic enzymes of the intrinsic coagulation cascade, i.e., factors IXa, Xa, Xla, and Xlla, also are neutralized by antithrombin III in a similar heparin-dependent way. However, heparin induces only a 4- to 15-fold enhancement in the rate of neutralization of these factors by antithrombin III.

Recent studies indicate that a very small percentage of the total HSPG synthesized by aortic and microvascular endothelial cells (∼1% and 10%, respectively) accounts for the majority of antithrombin III binding and anticoagulant activity. This HSPG binds to hydrophobic resins consistent with a membrane location and contains heparan sulfate chains that are enriched in the antithrombin III binding disaccharide sequence of GlcA—GlnNAc—3-O-SO3. These findings also demonstrate that molecules synthesized by cells other than mast cells possess anticoagulant properties.

It appears that endothelial surface proteoglycans are capable of binding not only anticoagulant factors but also procoagulant factors. For example, thrombin binds to the surface of cultured porcine aortic endothelial cells in a rapid and reversible manner and this binding is partially inhibited by pretreatment of the cells with purified heparitinase, an enzyme that cleaves heparan sulfate chains. Thus, it may be that the inactivation of thrombin by antithrombin III is regulated, at least in part, by one or more HSPGs present at the surface of the endothelial cell and that bind both coagulant and anticoagulant factors.

The role of platelets in hemostasis, thrombosis, and atherosclerosis is well established. Less clear is what specific role(s) platelet secretory products play in arterial wall metabolism. For example, platelets contain a variety of substances that are stored in alpha granules and released when platelets are stimulated to aggregate in tissue injury, promoting coagulation at the damaged site. Among these substances are a number of antiheparin factors such as platelet factor 4 (PF4), which binds to heparin and neutralizes heparin activity. PF4 occurs complexed to a chondroitin sulfate proteoglycan, which appears to function as a carrier molecule. More recent studies indicate that megakaryocytes synthesize at least two forms of CSPG and these forms, which are present within α granules of platelets, are released in response to thrombin. The significance of the PF4:PG complex in arterial wall metabolism needs to be elucidated. The fact that heparin releases PF4 into the circulation similar to the release of lipoprotein lipase suggests that PF4 also binds to a heparin-like molecule on the endothelial cell surface. In fact, additional studies demonstrate that PF4 binds to the surface of cultured human umbilical vein endothelial cells in a time-dependent, saturable fashion and that heparin and HS compete for binding.

Lipid Metabolism

Endothelial cell surface HSPGs also are involved in lipid metabolism. For example, lipoprotein lipase [a dimeric enzyme responsible for the hydrolysis of di- and triacylglycerol constituents of plasma very low density lipoproteins (VLDL) and chylomicrons] interacts with the surface of endothelial cells in a heparin/heparan sulfate-dependent fashion. Recently, Klinger et al. used a lipoprotein lipase-agarose affinity column to isolate an HSPG from a mixture of proteoglycans in rat brain and found it to be a membrane-associated proteoglycan of approximately 220 kD containing glycosaminoglycan chains of M, = 14 000 to 15 000. The mechanism by which this proteoglycan interacts with the enzyme is not clear, but most probably involves the glycosaminoglycan portion of the molecule, since glycosaminoglycan chain length and the number of N-sulfate and N-acetyl groups on the glycosaminoglycan chain affect affinity for lipoprotein lipase.

Another way in which endothelial cell-derived proteoglycan may modulate lipid metabolism is by modifying the charge density of the lipid as it passes through the endothelial barrier. The ability of cultured endothelial cells to bind, transport, and degrade low density lipoproteins (LDL) is well documented. Further studies indicate that LDL can be modified by endothelial cells so that it becomes more negatively charged, allowing recognition by the modified LDL receptor of the macrophage. It may be that endothelial-derived proteoglycans bind to LDL, and in turn, induce such a modification.

It is clear that regions of blood vessels that accumulate proteoglycans have a high propensity to accumulate lipid. For example, studies using the balloon injury model of experimental atherosclerosis indicate that proteoglycans accumulate within regions of injured vessels characterized by endothelial regrowth and not within regions devoid of endothelium. The role of proteoglycans in the re-endothelialized intima, the region characterized by excessive proteoglycan accumulation and not in the de-endothelialized portion (Figure 9). These studies also indicate that if animals are put on a high-fat diet, lipid accumulates in the re-endothelialized intima, the region characterized by excessive proteoglycan accumulation and not in the de-endothelialized portion (Figure 9).

Such studies raise the interesting possibility that during healing, the regenerating endothelium contributes to the proteoglycan composition of the arterial wall and in some way influences the metabolism of proteoglycans and lipids present in this region of blood vessel.

A number of studies now demonstrate that regions of vessels that have re-endothelialized have higher rates of proteoglycan synthesis and deposition than do de-endothelialized vessels. Co-culture experiments
work suggests that multiple pools of LDL-PG complexes may be present within blood vessels. For example, complexes extracted with saline contained chondroitin-6-sulfate as the major and hyaluronic acid as the minor glycosaminoglycan and were cholesterol ester-enriched. On the other hand, LDL-PG complexes isolated after collagenase treatment contained mostly hyaluronic acid and minor amounts of chondroitin-6-sulfate. Complexes isolated after elastase contained only hyaluronic acid and were cholesterol ester-poor. These findings suggest that particular LDL-PG complexes may occur within different regions of the extracellular matrix within blood vessels.

Camejo and his colleagues have been studying the interaction of arterial proteoglycan with lipoprotein and have found that arterial CSPG exhibits a marked affinity for LDL and that this interaction may be dependent not only on the type of proteoglycan but also on the surface charge of the LDL. For example, a recent study has shown that a specific sequence enriched in positively charged amino acids in the apo B moiety of LDL effectively binds to proteoglycans. These binding domains are part of the protein that interacts with LDL receptor. Thus, interaction of the proteoglycan with LDL may affect recognition of LDL by its surface receptor. In addition to CSPG, the small arterial DSPG also is capable of forming insoluble complexes with LDL in the presence of calcium, while those proteoglycans containing HS exhibit only minimal lipoprotein binding activities.

Intact proteoglycan is much more effective in precipitating LDL and VLDL than are isolated glycosaminoglycan chains. Removal of the protein core after β-elimination or protease treatment of the proteoglycan abolishes its insoluble complex-forming ability. The role of net negative charge in complex formation is emphasized by the observation that desulfation of the proteoglycan drastically reduces its ability to interact with LDL. Thus, it appears that PG-lipoprotein formation involves a complex series of multiple interactions of protein-protein and protein-carbohydrate. It also is possible that lipids influence the proteoglycan content of the vascular wall and, in turn, influence lipid deposition. Hoff and Wagner demonstrated that after 11 weeks on a hypercholesterolemic diet, porcine aortic glycosaminoglycan concentrations did not differ from that of normocholesterolemic animals, even though lipoproteins accumulated within the aorta. However, the relative amounts of aortic chondroitin sulfate increased, and dermatan sulfate decreased in the hypercholesterolemic animals, indicating that cholesterol feeding may influence the types of proteoglycans that accumulate but not the total quantity. Intense immunostaining using an anti-aortic CSPG antisera, has been observed in atherosclerotic lesions in the rabbit after lipoid feeding (Figure 3). The mechanism responsible for this specific accumulation is unclear, but it may be that lipoproteins influence the synthesis of proteoglycans by the resident cells of the arterial wall. LDL increase the synthesis of proteoglycans by cultured bovine smooth muscle cells, but it is unclear whether specific types of proteoglycans synthesized are affected.

The above studies demonstrate that proteoglycans within the extracellular matrix of blood vessels are capable of binding lipoproteins. However, lipid and lipoprotein

Figure 9. Correlation of lipid and proteoglycan concentration in de-endothelialized and re-endothelialized aortas in rabbits on normal and lipid-rich diets. Note that the re-endothelialized unstained (white) regions contained significantly more PG than the blue regions in animals on regular diets suggests that this increase in PG may predispose the white region to lipid accumulation.
are found not only within the extracellular matrix, but also within cells of the arterial wall during the genesis of the atherosclerotic lesion. Two cell types that are known to accumulate lipid in developing atherosclerotic lesions are smooth muscle cells and macrophages.102 and recent studies suggest that proteoglycans may play a role in intracellular lipid accumulation in these cells as well. For example, it is known that macrophages contain few receptors for LDL, and LDL will not accumulate within macrophages when these cells are incubated with high concentrations of LDL.242 However, if LDL is modified by acetylation, acetoxycylation, or malondialdehyde derivation, macrophages dramatically increase their uptake of modified LDL. Enhancement of LDL uptake also occurs when macrophages are incubated with medium containing dextran sulfate.243 These findings suggest that charge alteration of LDL molecules influences their recognition by macrophages. Thus, overall charge density of lipoproteins could be affected by complexing with negatively charged proteoglycans. Falcone et al.244 demonstrated that insoluble complexes of LDL, heparin, fibronectin, and collagen were taken up more rapidly in combination and to a greater extent by macrophages than when these cells were incubated with LDL alone. Furthermore, this study demonstrated that catabolism of the endocytosed LDL-complex was greatly diminished, causing cholesterol ester to accumulate within these cells. More recently, Hurt and Camejo245 demonstrated that incubation of human LDL with human arterial CSPG increased LDL uptake by human monocyte-derived macrophages, indicating that proteoglycan alone is capable of modifying LDL. It is interesting that sulfated polyanions such as dextran sulfate inhibit the fusion of phagosomes with lysosomes within macrophages by modifying membrane fluidity of the lysosome.246,247 This finding may partially explain why LDL is not degraded and accumulates in macrophages, leading to foam cell formation. However, decreased degradation of LDL may not be the only mechanism by which lipoprotein accumulates in macrophages. Salisbury et al.248 demonstrated increased cholesterol ester synthesis when macrophages were incubated with aortic proteoglycan and plasma LDL. In addition, Vijayagopal et al.249 showed that LDL complexed to aortic proteoglycan-hyaluronic acid aggregate was taken up by macrophages, but degraded more rapidly than LDL complexed to proteoglycan monomer. This enhanced degradation also was accompanied by increased cholesterol ester synthesis by the macrophage. This study also showed that acetyl-LDL completely inhibited the degradation of the LDL-PG complex, indicating that the complex may be recognized by the modified LDL receptor of the macrophage.

Atherosclerosis

Studies continue to demonstrate that proteoglycans accumulate within intimal lesions in both large and small vessels involved in atherosclerotic development. These increases appear to involve mainly the CSPG and DSPG families, while some studies indicate little change or even a decrease in HSPG with advancing atherosclerosis (for reviews see Hanspers 3, 4, 5, and 250 to 253). Little is known about whether the structural properties of intact proteoglycans are altered in developing atherosclerotic plaques. Wagner et al.34,254 noted that CSPG isolated from human fatty-fibrous plaques exhibited less tendency to form aggregates with hyaluronic acid and that the aggregates present in fatty-fibrous arterial plaques were smaller than those present in normal aorta. Altered aggregate formation may have some bearing on arterial calcification (a complication of advanced atherosclerotic plaques) since the degree of proteoglycan aggregation is thought to influence calcification.255 In addition, the CSPG isolated from human arterial lesions tends to be of larger hydrodynamic size, suggesting either that the synthesis or degradation of this monomer is altered or that it is complexed to other components in arterial lesions. Other minor changes in the structure of arterial proteoglycan have been noted. For example, DSPG isolated from atherosclerosis-susceptible pigeons contain glycosaminoglycans that are smaller than similar chains isolated from aortic DSPGs of pigeons that are less susceptible to this disease.256 Whether these differences influence the functional properties of arterial proteoglycans and whether other differences exist must await further investigation.

Conclusion

This review has concentrated on the role that proteoglycans play in vascular wall biology. As summarized in Figure 10, there are a number of ways in which proteoglycans can influence the behavior of vascular cells and, in turn, the normal and pathophysiological properties of blood vessels. Clearly, these macromolecules are of enormous importance in maintaining the viscoelastic state of the vessels, and any change in proteoglycan structure, content, or concentration will severely affect the biophysical properties of the tissue. We now know that a number of factors contribute to alteration of the proteoglycan content of the vascular wall through processes common to both normal development and disease, such as cell proliferation and migration, hormonal stimulation, and multiple ligand interactions. Such “alterations” may, in turn, generate conditions in which different proteoglycans acquire the capacity to “instruct” or to serve as important “informational transducers” regulating key events in vascular wall biology. The future of research in the cell biology of arterial proteoglycans is, indeed, challenging. It represents a critical component in the effort to understand those factors that contribute to cardiovascular disease—the leading cause of death in the United States and Europe.

Acknowledgments

I thank a number of colleagues who provided much of the original research reported in this review. Among them are Michael Kinsella, Susan Potter-Perigo, Michael Lark, Tet Kin Yeo, Tia Aulinskas, Sentaro Kobayashi, Alan Snow, Arlene Wechezak, Jacques Garrigues, Hans Krasse, Alex Clowes, and Linde Sandell. I also express sincere appreciation to Lakshmi Subbiah, Stephanie Lara, Kathy Braun, Henderson Mar, and Stephen MacFarlane for their technical expertise in studies reported from my laboratory. I also thank the many students who have worked on the various projects and who have stimulated our thinking. I am indebted to Anita Coen for the typing and editing of this manuscript.
Figure 10. A schematic representation of some of the possible roles that proteoglycans play in the biology of the arterial wall.

Functions

1. Binding of coagulation and anticoagulation factors
2. Binding and regulation of enzyme (lipoprotein lipase) activity
3. Carrier molecule for certain platelet products and plasma proteins
4. Binding and regulation of growth factor activity
5. Influencing cell-cell associations
6. Influencing cell adhesion
7. Participating in the organization of ECM structures such as basement membranes and regulating permeability
8. Influencing endothelial cell migration and proliferation
9. Modulation in arterial smooth muscle cell proliferation and migration
10. Regulation of collagen fibrillogenesis
11. Maintenance of viscoelastic properties
12. Modulating calcification
13. Influencing intra- and extracellular lipid deposition and turnover

References


55. Kinse IKA, Wight TN. Endothelial cell derived dermatan sulfate proteoglycan (DSPG) is croslinked to fibronectin by transglutaminase. J Biol Chem (in press)


58. Sakashita S, Engvall E, Ruoslahti E. Basement mem-

brane glycoprotein laminin binds to heparin. Fed Eur Bio-


60. Kanwar YS, Linker A, Farquhar MG. Increased perme-


68. Joyner WL, Mayhan WG, Johnson RL, Phares CK. Microvascular alterations develop in Syrian hamsters after the induction of diabetes mellitus by STZ. Diabe-

tes 1981;30:93–100


72. Chang Y, Yanagisawa M, Hascall VC, Wight TN. Proteo-
glycans synthesized by smooth muscle cells derived from monkey (Macaca nemestrina) aorta. J Biol Chem 1983;258:5679–5688


84. Wagner WD, Salisbury BGJ, Rowe HA. A proposed structure of chondroitin 6-sulphate proteoglycan of human normal and adjacent atherosclerotic plaque. Arteriosclero-
sis 1986;6:407–417


90. Hedman K, Christner J, Jullkunen I, Vaheri A. Chondroitin sulphate at the plasma membranes of cultured fibro-


91. Lark MW, Wight TN. Modulation of proteoglycan metabo-


Wechezak AR, Wight TN, Viggers RF, Sauvage LR. Endothelial adherence under shear stress is dependent upon microfilament re-organization. J Cell Physiol (in press)


Funderburg FM, Markwald RR. Conditioning of native substrates by chondroitin sulfate proteoglycans during cardiac mesenchymal cell migration. J Cell Biol 1986; 112:2475-2487


178. Deuel TF, Huang JS, Proffitt RT, Baenziger JJ, Chang D, Kennedy BB. Human platelet-derived growth factor...


188. Shimada K, Ozawa T. Evidence that cell surface heparan sulfate is involved in the high affinity thrombin binding to cultured porcine aortic endothelial cells. J Clin Invest 1985;75:1308–1316


Index Terms: proteoglycans • atherosclerosis • endothelial cell • smooth muscle cell • cell biology • hemostasis • proliferation • cell adhesion and migration • lipid-proteoglycan interactions
Cell biology of arterial proteoglycans.

T N Wight

doi: 10.1161/01.ATV.9.1.1

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://atvb.ahajournals.org/content/9/1/1