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Commentary

Vascular Endothelium, Hemodynamic Forces, and Atherogenesis

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Intimal lipid accumulation, hyperplasia, and scarring are stigmata of atherosclerotic vascular disease, whose major complications—myocardial and cerebral ischemia and infarction—continue to be major health problems in developed nations.[1] This insidiously progressive disease typically spans decades, but can reach a clinical horizon in a matter of minutes due to critical changes in a given atherosclerotic plaque that result in localized but life-threatening thrombosis. Epidemiological studies have established that hypercholesterolemia is an important risk factor in this disease process, and lipid-lowering drugs have been proven to have clinical efficacy. Experimental animals that are fed lipid-rich diets to elevate their plasma cholesterol levels also can develop atherosclerotic-like lesions, as do animals with naturally occurring or genetically engineered mutations that result in altered cholesterol metabolism. However, regardless of a given patient's risk factor profile, species of animal model, or type of natural or engineered genetic alteration, the early, lipid-rich lesions of atherosclerosis show a markedly nonrandom pattern of distribution within the arterial vasculature. Atherosclerotic lesions typically develop in the vicinity of branch points and areas of major curvature. These arterial geometries are associated with blood flow disturbances such as nonuniform laminar flow with boundary layer separation, complex secondary flows with flow reversal and dynamic stagnation points, and resultant temporal and spatial gradients in wall shear stresses. In contrast to these atherosclerosis-prone areas, unbranched, tubular arterial geometries, which are associated with a more uniformly laminar flow profile, characteristically are relatively atherosclerosis-resistant, at least in the early phases of the disease. This strikingly localized pattern of lesion formation, even in the face of systemic risk factors such as elevated plasma cholesterol, has intrigued experimental pathologists and fluid mechanical engineers alike for decades, and has motivated the search for a mechanistic link between hemodynamic forces and atherogenesis.

In this issue of *The American Journal of Pathology*, Zand and coworkers[2] describe a novel experimental model system for creating flow disturbances in the aorta of the rat that have significant effects on the pattern of intimal lipid deposition induced by chronic dietary hypercholesterolemia. Surgical insertion of a hemispherical glass plug into the aortic lumen, through the ostium of a severed renal artery, created a significant stenosis (greater than 50% cross-sectional area reduction) without causing any compression of the adjacent aortic wall, as typically occurs in other models involving an externally applied ligature or metal clip. This model thus accomplishes two useful things: it reliably creates an altered vascular geometry designed to induce well characterized intraluminal flow perturbations while it minimizes the confounding issue of concomitant (and often less well characterized) changes in intramural stresses and strains. Detailed quantitative studies of the flow field perturbations created by the hemispherical plug were performed in parallel, using a scaled-up *in vitro* biophysical model. Histopathological analysis of the experimental aortas showed a clear-cut association of intimal lipid deposition with certain types of flow perturbation. Crescentic areas of intimal lipid accumulation and subendothelial thickening were localized both proximal and especially distal to the plug, in the regions of predicted low wall shear stress, flow separation, stagnation, and recirculation. In contrast, there were essentially no lipid deposits visible in the intimal area opposite the plug, ie, in a region of increased wall shear stress. These observations, in particular the asymmetric expansion of intimal changes in the direction of flow, also may have implications for the growth of

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raised atherosclerotic lesions. As the authors point out, an association of intimal lipid deposition with disturbed flow has been inferred previously by others studying the focal nature of atherosclerotic lesions in various experimental animals and human subjects. However, the current study is perhaps the clearest demonstration, to date, of this phenomenon, experimentally generated in a biophysically well characterized *in vivo* model.

What are the implications of these observations for the pathogenesis of atherosclerosis? In particular, what mechanisms at the cellular and molecular level link hemodynamic forces with vessel wall pathophysiology? As early as 1969, Caro and coworkers[3] called attention to the association of low shear areas with early atherosclerotic lesions in human subjects. This led to the hypothesis that low (time-averaged) wall shear stresses, such as occur near the carotid bifurcation and other atherosclerosis-prone geometries, might result in prolonged resident times for large, atherogenic lipid particles (eg, low-density lipoproteins) or even blood cells (eg, platelets, mononuclear leukocytes), thus favoring their localized attachment and infiltration into the arterial wall. This reasoning also can be extended to the effect of low wall shear on local boundary-layer, mass transport-dependent processes, resulting in the localized accumulation of cytokines, growth factors, or reactive oxygen species in the vicinity of the intimal surface. This working concept invokes rheological issues per se as the critical determinants and views the contribution of the vessel wall as relatively passive. In the last two decades, however, there has been a major paradigm shift in our mechanistic thinking in vascular biology—one that places a primary emphasis on the active involvement of the cells that comprise the arterial wall, in particular endothelium and smooth muscle, in vascular disease processes such as atherosclerosis.[1,4] In this context, an alternative hypothesis linking hemodynamic forces and atherosclerosis has emerged, one which already has generated a number of novel mechanistic insights. The central premise of this new working hypothesis is that the endothelial lining, as the cellular component in direct contact with flowing blood, is the primary sensor of wall shear stresses and functions as a transducer of these biomechanical stimuli into biological responses within the vessel wall.[5–7]

Inferential evidence that vascular endothelial cells *in vivo* are responsive to local flow conditions comes from *en face* morphological analyses of cell shape in different regions of the arterial intima. In areas of uniform laminar flow, endothelial cells exhibit an ellipsoidal cell (and nuclear) shape and alignment in the direction of flow, whereas in regions of disturbed flow, this orderly pattern is disrupted.[8,9] In addition, arterial wall remodeling (eg, after experimental surgical coarctation or shunting procedures) appears to depend at least in part on a functionally intact endothelium.[10] Direct evidence that hemodynamic forces can influence endothelial structure and function has come from studies in which cultured endothelial cells have been subjected to defined fluid mechanical forces under well controlled conditions *in vitro*. In these simplified model systems, unidirectional steady laminar shear stresses of physiological amplitude induce time- and force-dependent endothelial cell shape change and alignment that is gradually reversible upon the cessation of flow.[11–13] These shear-induced changes in cell shape are accompanied by reorganization of actin filaments and other cytoskeletal components, thus mimicking the morphology of aortic endothelium *in vivo*. Additionally, work by numerous laboratories has demonstrated a variety of changes in the metabolic and synthetic activities of endothelial cells in response to defined flow stimulation, including the production of prostacyclin, nitric oxide, cytokines, growth factors, extracellular matrix components, and vasoactive mediators.[5–7,14] Some of the more acute changes involve regulation at the level of rate-limiting enzymes and/or substrate availability (eg, arachidonic acid release by calcium-sensitive phospholipases, NO production by nitric oxide synthase). However, in many cases where the responses are delayed and/or sustained, modulation of endothelial gene expression appears to be occurring.

How is the frictional force of blood flow along its luminal surface sensed by the endothelial cell and transduced into molecular biological events such as gene regulation? At present, the fundamental question of the identity, location, and mechanism(s) of action of endothelial flow-sensitive mechanotransducers remains a challenging one. Several distinct molecules, eg, cell surface ion channels, various receptor-associated G proteins, and members of the mitogen-activated and stress-activated protein kinase cascades, as well as certain transcription factors such as nuclear factor-κB, c-Fos, and Egr-1, are rapidly activated in response to fluid shear stresses applied to the endothelial cell surface.[5,6,14–17] In addition, cellular organelles, such as the cytoskeleton (intermediate filaments, microtubules, actin-myosin stress fibers), plasma membrane caveoli, lateral cell-cell junctional proteins, basal focal adhesion complexes, and even the lipid bilayer of the plasma membrane, also appear to be participating in shear-induced endothelial responses. Finally, various second messengers, including ionized cytosolic calcium, intracellular lipid products of the polyphosphoinositide pathway, and nitric oxide, are generated in the context of flow-stimulation of endothelium. As discussed by Davies,[5] the challenge is to understand the interaction of these spatially and temporally disparate components in the dynamic interplay of the endothelial cell's response to biomechanical stimulation—sorting out where transmission becomes transduction, as well as cause-effect relationships. For example, one might envision the endothelial cell as analogous to a circus tent and the wind blowing across the roof of the tent as the frictional force (wall shear stress) generated by blood flow. As the wind blows, the ropes tethering the tent roof (analogous to cytoskeletal stress fibers) transmit fluctuating forces to the stakes (analogous to integrin-containing focal adhesion complexes) anchored in the ground around the base of the tent. The resulting movement of the stakes in the earth might be likened to phosphorylation/dephosphorylation and other biochemical events occurring in the focal adhesion complexes distributed along the basal aspect of the cell, which then are functioning as biomechanical-biochemical transducers

at a distance from the point of application of the force. Similar actions at a distance might be envisioned between apical membrane and lateral cell-cell junctions, and/or the nuclear envelope.[5] Indeed, as Ingber[18] has suggested, the entire cytoskeleton may participate in what he called a tensegrity-based stimulus-response coupling, which dynamically integrates externally applied stresses, extracellular matrix attachments, and internal strains into adaptive biological responses. Given that the endothelial cell cytoskeleton and focal adhesion complexes can actively be remodeled in response to applied forces[5] and that there are regional differences in the magnitude of shear stresses across the surface of a given endothelial cell (reflecting details of its surface topography),[19,20] the potential for significant heterogeneity in responsiveness among cells within a uniform flow field also exists. Interestingly, although this is a relatively complex model for biomechanical transduction, considerable observational data support it. In contrast, at present there is no clear-cut example of an intrinsic membrane protein whose deformation by shear stresses directly results in transmembrane signal transduction, ie, a candidate shear receptor.

Regardless of the proximal sensing mechanism(s), how does a given gene within the endothelial nucleus become transcriptionally activated in response to shear stress? Using the human platelet-derived growth factor-B chain (PDGF-B) as a model of a shear-responsive gene in endothelial cells, Resnick and coworkers[21] defined the first example of a "shear stress response element (SSRE)." This SSRE, consisting of a simple 6-bp (GAGACC) motif in the proximal promoter region, was shown to function as a necessary and sufficient *cis*-acting transcriptional regulatory element, mediating the up-regulation of PDGF-B expression in response to physiological levels of applied fluid shear stresses. Subsequent studies[22] established that components of the NF-κB complex could interact functionally with this SSRE to mediate transcriptional activation of the PDGF-B gene. Previous work had established that NF-κB was rapidly activated, as evidenced by its translocation to the nuclear compartment, in response to physiological levels of laminar shear stress (LSS). Taken together, these studies thus established the first model for coordinate *trans*-activating and *cis*-activating transcriptional regulation in response to biomechanical stimulation in the endothelial cell. Subsequent studies have established the existence of various other SSREs in several pathophysiologically relevant endothelial genes (eg, monocyte chemoattractant protein-1, tissue factor, endothelin-1, VCAM-1) which appear to be at least partly responsive to biomechanical stimulation. Interestingly, in the case of VCAM-1, the relevant SSRE appears to mediate the down-regulation of transcription in response to LSS stimulation.[23] Indeed, recent studies[24] have shown that LSS stimulation can blunt the up-regulation of VCAM-1 in endothelial cells activated by various proinflammatory cytokines. These observations may have special relevance to the basic question of atherosclerotic lesion localization *in vivo*, as will be considered in greater detail below. Some further points should be noted with regard to SSREs and shear stress gene regulation in endothelial cells. Many genes contain one or more copies of the various SSREs identified to date, yet not all of these genes are necessarily shear stress-responsive. Indeed, SSREs appear to function in a context-specific manner, in that a given element can mediate transcriptional responses to shear stress in the context of one promoter and yet not do so in the context of another promoter.[14] This has been illustrated in studies of LSS induction of leukocyte-endothelial adhesion molecules, including E-selectin, ICAM-1, and VCAM-1, each of which contains functional NFκ-B elements but responds differently (ICAM-1, up-regulation; VCAM-1, no change or down-regulation; E-selectin, no change) in the same cell exposed to the same biomechanical stimulus.[25,26] It should also be noted that certain transcription factor genes, eg, Egr-1 and c-fos, are themselves shear-responsive,[27] thus raising the possibility that they may function in the induction of multiple other genes in the orchestration of an endothelial cell's response to biomechanical stimulation.

Do differences in the temporal and/or spatial characteristics of flow elicit different patterns of endothelial gene expression? Studies by several laboratories using different *in vitro* model systems clearly indicate that endothelial cells can sense differences in the temporal and/or spatial characteristics of flow and translate these biomechanical stimuli into different biological responses. For example, steady laminar flow appears to enhance endothelial cell survival by suppressing apoptosis,[26,28] whereas turbulent flow can trigger endothelial cell division.[29] Differences in the temporal properties of laminar flow stimulation, generated by instantaneous (impulse) *versus* gradual (ramp) application of the same final level of shear stress, can elicit very different responses in endothelial gene expression.[30,31] Similarly, oscillatory *versus* steady laminar flows elicit marked differences in the pattern of adhesion molecule expression in cultured endothelium.[32] To focus more specifically on the effects of spatial gradients in shear stress on endothelial biology, DePaola et al[33] developed an *in vitro* model system that generates large gradients in shear stress over the relatively small dimensions of a cultured endothelial monolayer, thus mimicking the spatial pattern of flow separation (with reversal), reattachment, and flow recovery associated with arterial bifurcations *in vivo*. Using this *in vitro* spatial disturbed flow model, dramatic differences in endothelial cell shape, migration, and proliferation have been demonstrated in association with disturbed flow as compared with uniform laminar flow.[34] In addition, significant differences in endothelial expression of Connexin43 at the level of mRNA and protein, and concomitant changes in cell-cell communication via gap junctions, also have been correlated with the presence of shear stress gradients in this model.[35] Recent studies suggest that these *in vitro* observations may indeed have a counterpart *in vivo*.[36] Most recently, Nagel and coworkers[37] have demonstrated that endothelial cell monolayers exhibit significant spatial heterogeneity in the nuclear localization of certain critical transcription factors, including NF-κB, Egr-1, c-jun, and c-fos, and that these differences correlate with the local shear stress gradient. Taken together,

these studies thus strongly suggest that spatial gradients of wall shear stress, in contrast to absolute shear stress magnitudes, can be important determinants of endothelial responses at the level of gene regulation. Further studies are needed to elucidate the potential interplay of both temporal and spatial fluctuations in the biomechanical regulation of endothelial gene expression and, ultimately, to correlate these stimuli with the endothelial phenotypes actually observed in different *in vivo* biomechanical environments.

How might flow-induced changes in endothelial phenotype contribute to atherogenesis *in vivo*? To more systematically address the question of modulation of endothelial phenotype by biomechanical stimuli, our laboratory has turned to high- throughput molecular biological strategies. Specifically, we have used a reverse transcription-polymerase chain reaction-based, high-throughput differential display of transcripts to compare the patterns of genes that are up-regulated or down-regulated in human endothelial cells in response to physiological levels of steady laminar shear stress, a comparable level of turbulent (nonlaminar) shear stress, and a soluble cytokine stimulus (interleukin-1β) at a maximally effective concentration.[14] This approach has revealed distinctive patterns of endothelial gene expression not previously appreciated, including a set of genes that appear to be up-regulated in a sustained fashion by steady laminar shear stress but not by turbulent shear stress. Certain of these differentially regulated transcripts encode known endothelial genes of relevance to atherogenesis, such as eNOS or NOSIII (the endothelial isoform of nitric oxide synthase), COX-2 (the inducible isoform of cyclooxygenase), and Mn-SOD (manganese-dependent superoxide dismutase). These endothelial genes encode enzymes that exert potent anti-thrombotic, anti-adhesive, anti-proliferative, anti-inflammatory, and anti-oxidant effects, both within the endothelial lining and in interacting cells such as platelets, leukocytes, and vascular smooth muscle. The biological consequences of these steady laminar shear up-regulated endothelial genes thus would be predicted to be vasoprotective or anti-atherogenic.[14,38]

Given that uniform laminar shear stresses are characteristically associated with atherosclerotic lesion-protected arterial geometries *in vivo*, these observations have led us to hypothesize that this type of biomechanical stimulus up-regulates the expression of a subset of "atheroprotective genes" in endothelial cells, which then act locally in the lesion-protected areas to offset the effects of systemic risk factors such as hypercholesterolemia, hyperhomocysteinemia, hyperglycemia (diabetes), and hypertension.[4,14,27] The coordinated and selective up-regulation of atheroprotective genes by uniform laminar shear stress is a possible mechanistic link between the local hemodynamic milieu, endothelial gene expression, and early events in atherogenesis, thus providing a potential explanation for the nonrandom localization of early atherosclerotic lesions. This working hypothesis does not exclude the potential direct action of complex disturbed laminar flows, such as occur in lesion-prone arterial geometries, as stimuli for the expression of pro-atherogenic genes (eg, adhesion molecules, growth factors, cytokines).[27]

Critical testing of the "atheroprotective gene" and related hypotheses will depend on refinement of both *in vitro* and *in vivo* fluid mechanical models, and their applicability to vascular disease pathobiology at the molecular genetic level. The development of reliable methods for the linear amplification of transcripts from small numbers of cells and their analysis by DNA chip microarrays or analogous genome scale technologies hold much promise in this regard. Application of these comprehensive and relatively unbiased methods of molecular analysis to endothelial cells subjected to experimentally defined flow conditions will add significantly to our understanding of the dynamic range of biomechanically induced phenotypic modulation. Ultimately, the extension of this approach to dysfunctional vascular endothelium in the natural disease context should provide new insights into the link between hemodynamic forces and atherogenesis.

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References

1. Ross R: Atherosclerosis: an inflammatory disease. N Engl J Med 1999, 340:115–126
2. Zand T, Hoffman AH, Savilonis BJ, Underwood JM, Nunnari JJ, Majno G, Joris IS: Lipid deposition in rat aortas with intraluminal hemispherical plug stenosis: a morphological and biophysical study. Am J Pathol 1999, 155:XXX-xxx
3. Caro CG, Fitz-Gerald JM, Schroter RC: Arterial wall shear and distribution of early atheroma in man. Nature 1969, 223:1159–1161
4. Gimbrone MA Jr, Topper JN: Biology of the Vessel Wall: Endothelium. Molecular Basis of Heart Diseases. Part V: Vascular Biology & Atherogenesis. Edited by KR Chien, JL Breslow, JM Leiden, RD Rosenberg, CE Seidman. Troy, MO, Harcourt Brace & Co, 1998, pp 331–348
5. Davies PF: Flow-mediated endothelial mechanotransduction. Physiol Rev 1995, 75:519–560
6. Resnick N, Gimbrone MA Jr: Hemodynamic forces are complex regulators of endothelial gene regulation (review). FASEB J 1995, 9:874–882
7. Gimbrone MA Jr, Nagel T, Topper JN: Biomechanical activation: an emerging paradigm in endothelial adhesion biology. Perspectives Series: Cell Adhesion in Vascular Biology. J Clin Invest 1997, 99: 1809–1813
8. Reidy MA, Bowyer DE: Scanning electron microscopy of arteries: the morphology of aortic endothelium in haemodynamically stressed areas associated with branches. Atherosclerosis 1977, 26:181–194
9. Levesque MJ, Liepsch D, Moravec S, Nerem RM: Correlation of endothelial cell shape and wall shear stress in a stenosed dog aorta. Arteriosclerosis 1986, 6:220–229
10. Langille BL, O'Donnell F: Reductions in arterial diameter produced by

chronic decreases in blood flow are endothelium-dependent. Science 1986, 231:405–407
11. Dewey CF Jr, Bussolari SR, Gimbrone MA Jr, Davies PF: The dynamic response of vascular endothelial cells to fluid shear stress. J Biomech Eng 1981, 103:177–185
12. Levesque MJ, Nerem RM: The elongation and orientation of cultured endothelial cells in response to shear stress. J Biomech Eng 1985, 107:341–347
13. Remuzzi A, Dewey CF, Davies PF, Gimbrone MA Jr: Orientation of endothelial cells in shear field in vitro. Biorheology 1984, 21:617–630
14. Topper JN, Gimbrone MA Jr: Blood flow and vascular gene expression: fluid shear stress as a modulator of endothelial phenotype. Mol Med Today 1999, 5:40–46
15. Malek AM, Izumo S: Molecular aspects of signal transduction of shear stress in the endothelial cell (review). J Hypertension 1994, 12:989–999
16. Traub O, Berk BC: Laminar shear stress: mechanisms by which endothelial cells transduce an atheroprotective force. Arterioscler Thromb Vasc Biol 1998, 18:677–685
17. Ohno M, Cooke JP, Dzau VJ, Gibbons GH: Fluid shear stress induces endothelial transforming growth factor β-1 transcription and production: modulation by potassium channel blockade. J Clin Invest 1995, 95:1363–1369
18. Chicurel ME, Chen CS, Ingber DE: Cellular control lies in the balance of forces. Curr Opin Cell Biol 1998, 10:232–239
19. Davies PF, Mundel T, Barbee KA: A mechanism for heterogeneous endothelial responses to flow in vivo and in vitro. J Biomech 1995, 28:1553–1560
20. Satcher RL Jr, Bussolari SR, Gimbrone MA Jr, Dewey CF Jr: The distribution of fluid forces on model arterial endothelium using computational fluid dynamics. J Biochem Eng 1992, 114:309–316
21. Resnick N, Collins T, Atkinson W, Bonthron DT, Dewey CF Jr, Gimbrone MA Jr: Platelet-derived growth factor B chain promoter contains a cis-acting fluid shear-stress-responsive element. Proc Natl Acad Sci USA 1993, 90:4591–4595
22. Khachigian LM, Resnick N, Gimbrone MA Jr, Collins T: Nuclear factor-kB interacts functionally with the platelet-derived growth facor B-chain shear-stress-response-element in vascular endothelial cells exposed to fluid shear stress. J Clin Invest 1995, 96:1169–1175
23. Korenaga R, Ando J, Kosaki K, Isshiki M, Takada Y, Kamiya A: Negative transcriptional regulation of the VCAM-1 gene by fluid shear stress in murine endothelial cells. Am J Physiol 1997, 273:C1506–C1515
24. Varner SE, Nerem RM, Alexander RW, Medford RM: Differential suppression of inducible endothelial VCAM-1 gene expression and NF-κB activation by laminar shear stress. Ann Biomed Engr 1996, 24:S56 (abstr.)
25. Nagel T, Resnick N, Atkinson WJ, Dewey CF J, Gimbrone MA Jr: Shear stress selectively upregulates intercellular adhesion molecule-1 expression in cultured human vascular endothelial cells. J Clin Invest 1994, 94:885–891
26. Garcia-Cardena G, Anderson KR, Mauri L, Gimbrone MA: Distinct mechanical stimuli differentially regulate the P13K/Akt survival pathway in endothelial cells. Proceedings of the Fifth Saratoga International Conference on Atherosclerosis. Ann NY Acad Sci 1999, in press
27. Topper JN, Gimbrone MA Jr: Hemodynamics and endothelial phenotype: new insights into the modulation of vascular gene expression by fluid mechanical stimuli. Endothelium and Mechanical Forces. Edited by P Lelkes. London, Harwood Academic Publishers, 1999, in press
28. Dimmeler S, Haendeler J, Rippmann V, Nehls M, Zeiher AM: Shear stress inhibits apoptosis of human endothelial cells. FEBS Lett 1996, 399:71–74
29. Davies PF, Remuzzi A, Gordon EJ, Dewey CF Jr, Gimbrone MA Jr: Turbulent fluid shear stress induces vascular endothelial cell turnover in vitro. Proc Natl Acad Sci USA 1986, 83:2114–2117
30. Hsieh HJ, Li NQ, Frangos JA: Pulsatile and steady flow induces c-fos expression in human endothelial cells. J Cell Physiol 1993, 154:143–151
31. Frangos JA, Bao XP: NO regulated PDGF-A and MCP-1 expression induced by temporal gradients in shear stress in human endothelial cells. Jpn J Pharmacol 1997, 75:31
32. Chappell DC, Varner SE, Nerem RM, Medford RM, Alexander RW: Oscillatory shear stress stimulates adhesion molecule expression in cultured human endothelium. Circ Res 1998, 82:532–539
33. DePaola N, Gimbrone MA Jr, Davies PF, Dewey CF Jr: Vascular endothelium responds to fluid shear stress gradients. Arterioscler Thromb 1992, 12:1254–1257
34. Tardy Y, Resnick N, Nagel T, Gimbrone MA Jr, Dewey CF Jr: Shear stress gradients remodel endothelial monolayers in vitro via a cell proliferation-migration-loss cycle. Arterio Thromb Vasc Biol 1997, 17:3102–3106
35. DePaola N, Davies PF, Pritchard WF Jr, Florez L, Harbeck N, Polacek DC: Spatial and temporal regulation of gap junction Connexin43 in vascular endothelial cells exposed to controlled disturbed flows in vitro. Proc Natl Acad Sci USA 1999, 96:3154–3159
36. Gabriels JE, Paul DL. Connexin43 is highly localized to sites of disturbed flow in rat aortic endothelium but connexin37, and connexin40 are more uniformly distributed. Circ Res 1998, 83:636–643
37. Nagel T, Resnick N, Dewey CF Jr, Gimbrone MA Jr: Vascular endothelial cells respond to spatial gradients in fluid shear stress by enhanced activation of transcription factors. Arterio Thromb Vasc Biol 1999, in press
38. Topper JN, Cai J, Falb D, Gimbrone MA Jr: Identification of vascular endothelial genes differentially responsive to fluid mechanical stimuli: cyclooxygenase-2, manganese superoxide dismutase, and endothelial cell nitric oxide synthase are selecively up-regulated by steady laminar shear stress. Proc Natl Acad Sci USA 1996, 93: 10417–10422