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The Contribution of Vascular Smooth Muscle to Aortic Stiffness Across Length Scales

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Abstract

The operation of the cardiovascular system in health and disease is inherently mechanical. Clinically, aortic stiffness has proven to be of critical importance as an early biomarker for subsequent cardiovascular disease; however, the mechanisms involved in aortic stiffening are still unclear. The etiology of aortic stiffening with age has been thought to primarily involve changes in extracellular matrix protein composition and quantity, but recent studies suggest a significant involvement of the differentiated contractile vascular smooth muscle cells in the vessel wall. Here, we provide an overview of vascular physiology and biomechanics at different spatial scales. The processes involved in aortic stiffening are examined with particular attention given to recent discoveries regarding the role of vascular smooth muscle.

Keywords

aortic stiffness; focal adhesions; vascular smooth muscle cells; biomechanics

INTRODUCTION

Cardiovascular disease, the leading cause of death worldwide, is a group of complex, multifactorial disorders of the heart and blood vessels, including coronary heart disease, myocardial infarction, angina pectoris, heart failure, and stroke [18,38,81]. Traditional risk factors for cardiovascular disease include age, gender, hypertension, cholesterol, tobacco smoking, alcohol consumption, family history, diet, obesity, physical activity, and diabetes mellitus [18]. An additional independent risk factor—aortic stiffness—has recently attracted the renewed attention of clinicians and researchers. Aortic stiffening has been shown in numerous studies to precede and predict negative cardiovascular outcomes, but its development is not well understood [30]. As aortic stiffening is an early event in aging-associated cardiovascular diseases, often before the onset of symptoms, its study may provide clues to the underlying cellular and molecular mechanisms of cardiovascular disease and lead to novel therapeutic interventions. This review presents a brief summary of elements of cardiovascular physiology and biomechanics (reviewed in depth elsewhere [27,32,71]) that are important in the context of aortic stiffness. These elements are organized

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by biological length scale, from organ to subcellular levels (Figure 1), with particular focus given to the contribution of contractile vascular smooth muscle to aortic stiffness.

CARDIOVASCULAR PHYSIOLOGY

The Organ/System Level

The cardiovascular system functions to circulate blood throughout the body. The left ventricle of the heart pumps oxygenated blood through the successively smaller vessels of the bifurcating systemic arterial tree, through the proximal, large-diameter aorta, and subsequent elastic arteries, then the muscular arteries, and finally the distal arterioles, through the capillaries, where gas and nutrient exchange occur, and then through the venous tree [13,23]. The arteries most proximal to the heart are considered elastic conduits whose walls expand to accommodate the blood ejected by the heart and then recoil (Figure 1A), releasing their stored elastic energy and aiding in the propulsion of blood down the arterial tree. The flow of blood through the system is primarily impeded by the peripheral arteries of the microcirculation, which account for roughly 60% of the total resistance. By the time blood has reached the capillaries, the mean pressure has dropped substantially, the pulse pressure (the difference between systolic and diastolic blood pressures) has dampened out, and the flow is relatively steady.

The Tissue Level

The blood vessel wall, which is organized into three layers (Figure 1B), is composed of cells, extracellular matrix (principally the proteins collagen and elastin), and cell–cell and cell–matrix connections that link these components [5]. Blood vessels exhibit marked spatial heterogeneity between different wall layers and along the length of the vascular tree [5,21,22]. The outer adventitial layer contains fibroblasts, and the inner intimal layer contains endothelial cells. The endothelium produces potent vasoregulatory agents that modulate the tone of the dVSMCs found in the media, which is generally the thickest layer of the vessel wall. Smooth muscle contraction in microvessels can dramatically reduce lumen size, functioning to both increase vascular resistance and direct blood flow to tissues based on metabolic need. In large arteries the lumen reduction is modest, but stiffness is markedly increased in response to dVSMC contraction [5,19,23,55].

The Cellular/Subcellular Level—Vascular Smooth Muscle

Vascular smooth muscle contraction is initiated and regulated by various neurotransmitters, hormones, and paracrine factors, as well as mechanical stimulation and electrical stimulation, and a combination of these signals may be present at any given instant *in vivo*. Force generation derives from interactions between cytoskeletal proteins myosin and actin [23,33]. At least three kinds of pathways regulate vascular smooth muscle contractility: (i) those that regulate myosin activity; (ii) those that regulate actin availability; and (iii) those that regulate the nonmuscle actin cytoskeleton and FAs, integrin-based cell–matrix adhesions (traditionally called dense plaques or adhesion plaques in smooth muscle) (Figure 1C) [34,87]. This third regulatory scheme has come to light only recently from studies conducted mainly in airway smooth muscle [20,45,61,64,87] and runs contrary to classic models of a static ultrastructure for differentiated smooth muscle [5,60].

Although cytoskeletal plasticity is now recognized as an important factor in vascular smooth muscle as well, particularly in the cell cortex [6,34], most of what is known about FAs comes from studies of migrating cultured cells, where FA undergoes dynamic, tension-mediated formation, growth, and maturation, followed by release of the contacts with the matrix and recycling [9,14,28,78,84,88]. Tyrosine phosphorylation of FA proteins in migrating cells modulates FA size, location, and composition and is considered the biochemical signature of FA recycling and turnover [10,42,75,85]. FAs, which contain upward of 100 proteins, serve as the mechanical bridge between the outside of the cell and the actin cytoskeleton [15,16,37,49,86]. The FA performs mechanotransduction, wherein physical forces (hydrostatic pressure, shear, tensile stretch, etc.) are transformed into biochemical signals [4,53]. Aberrant mechanotransduction has been suggested to play a role in the pathogenesis and progression of cardiovascular and other diseases, but further investigation is necessary [29].

CARDIOVASCULAR BIOMECHANICS

The Organ/System Level

The nature of blood flow (pulsatile hemodynamics) is complex, determined not only by the time-variant pressure gradient established by contractions of the heart but also by the morphology (geometry), composition, and mechanical properties of the vessel walls, which vary along the length of the vascular tree [5,23]. Early attempts to model the arterial circulation and relate pressure to flow invoked the Windkessel model [46,76]. In its simplest form, this lumped parameter circuit model consists of a resistor representing the small arteries and microcirculation and a capacitor representing the compliance of the large arteries. This electrical analog captures the smoothing action of the aorta on flow from the heart, but incorrectly assumes that changes in pressure are felt instantaneously, without delays, throughout the entire system [40,41]. A more accurate model of the spatiotemporal variation in pressure and flow in the arterial tree treats the arterial system as a distributed load, using transmission line theory to account for transit delays, impedance mismatches between arterial segments, and wave reflections [2,40,65,66]. These models provide a valuable framework for describing and interpreting changes in cardiovascular function.

The Tissue Level

The biomechanical properties of the aorta wall depend on its material composition and organization. Vascular stiffness is commonly attributed to the major matrix proteins of the vessel wall, collagen, and elastin [71]. The basic notion is that at low pressures wall tension is born by distensible elastin, whereas at higher pressures the collagen fibers in the wall uncrimp and reorient to bear a greater portion of the load and effectively stiffen the vessel [26,52]. Elastin is responsible for the elastic recoil of the vessel in response to pulsatile flow, and the collagen protects the vessel from damage from overdistension.

The blood vessel wall exhibits mechanical behavior that is nonlinear, anisotropic (directionally dependent), and visco-elastic (exhibiting creep, stress relaxation, and hysteresis), with hysteresivity that is insensitive to strain rate [13]. In the unloaded state, the wall harbors residual stresses, and *in vivo* it exists in a prestretched, prestressed

configuration [13]. While many quantitative models have been proposed to characterize this complex biomechanical behavior, most only possess a subset of these features [70]. In particular, most models primarily describe passive mechanical properties with very few attempting to account for active cellular behaviors.

The Cellular/Subcellular Level—Vascular Smooth Muscle

The contribution of the contractile smooth muscle cell to aortic stiffening with age has largely been overlooked. The dVSMC is believed to act as a short-term tension setter that does not directly contribute to the stiffness of the vessel wall, but rather contracts transiently to redistribute tensile forces between elastin and collagen, modulating stiffness only on short timescales [44,51,80]. However, this scheme has been challenged by recent studies demonstrating for the first time that the intrinsic stiffness of aortic smooth muscle cells increases with either age or hypertension, through mechanisms tied to the actin cytoskeleton, and may contribute directly to aortic stiffening [50,56]. Further studies of the mechanical behavior of individual differentiated vascular smooth muscle cells are necessary to bridge the gap in our understanding between vessel mechanics at the micro- and macroscales [39].

AORTIC STIFFNESS

Functional vs. Material Aortic Stiffness

Currently, the literature and the field have taken to referring to the various measures of aortic stiffness simply as “aortic stiffness,” which is imprecise and may cause confusion. A preferred nomenclature would invoke two varieties of stiffness—material stiffness and functional (or effective) stiffness. Material stiffness (e.g., the modulus of elasticity or Young’s modulus E) is geometry independent, whereas functional stiffness (e.g., PWV, impedance, etc.) depends on vessel geometry (e.g., diameter, thickness, *in vivo* prestrain, and tethering) as well as the material composition, and therefore material stiffness, of the vessel wall [19].

Functional aortic stiffness is assessed by noninvasive clinical procedures *in vivo*, and some researchers have tried to estimate material stiffness from the *in vivo* data using mathematical relations derived from hemodynamics (e.g., the Bramwell–Hill or Moens–Korteweg equations) [44]. The gold standard of clinical measures is PWV, which is determined by recording carotid and femoral pressure waveforms and then dividing the transit distance by the transit time between the two sites [40]. It is important to clarify that PWV is not the velocity at which blood travels through the lumen, but rather the velocity at which the pressure signal is conducted by the vessel wall. Another surrogate for material stiffness is aortic characteristic impedance Z_c, a measure of the vessel’s capacity for limiting the flow induced by a time-varying pressure gradient [2,40]. Aortic PWV and characteristic impedance have the same dependence (square root) on Young’s modulus and thickness [40]. Importantly, PWV has an inverse square root dependence on lumen diameter, while characteristic impedance is fivefold more sensitive to changes in diameter [40]. This distinction is critical for proper interpretation of functional stiffness measurements, especially as there are situations in which PWV and characteristic impedance change divergently [40].

Although material stiffness can be calculated from noninvasive measures of functional stiffness, which reflect not only the material stiffness but also the geometry, strains, and pressurization of the vessel *in situ*, in practice there may be substantial errors introduced due to the challenges of monitoring and controlling the relevant geometric and loading parameters during blood flow *in vivo*. For this reason, the study of material stiffness is better suited to direct measurement of suitable animal tissues *in vitro* (e.g., by uniaxial stretching of rings as in [55], by biaxial stretching of tissue strips as in [68], or by pressurization of intact vessels as in [72]). In some regards such experiments will allow investigators to more quickly and easily understand the biomechanical and morphological underpinnings of cardiovascular disease.

Implications of Increased Functional Aortic Stiffness for Cardiovascular Disease

It is thought that the increased functional stiffness of the aorta with age decreases the impedance mismatch between the aorta and the muscular arteries, allowing greater transmission of the pulse pressure to distal sites [40]. This excess energy causes end-organ complications by damaging the microvasculature in critical high-flow, low-impedance organs, such as the brain, retina, and kidney [25,35,69,73]. The increased pressure pulsatility may also promote hypertrophy or remodeling in the resistance vessels, which would affect resistance to flow and the blood supply to various tissues and could lead to subsequent development of hypertension [40,43]. Changes in aortic stiffness may also shift the return timing of reflected waves to earlier in systole, which could augment aortic pressure and increase the load on the heart [41].

Mechanisms of Changes in Aortic Stiffness

Aortic stiffening with age or disease is generally thought to be a consequence of modifications to the morphology and composition of elastin and collagen [19,71]. In the aorta, repeated loading cycles over time lead to the fatigue failure of elastin and a progressive shift of mechanical loads from damaged, fragmented elastin fibers to collagen, thereby stiffening the vessel. Elastin has a relatively long half-life in the body (>40 years); however, essentially all elastin synthesis is complete by adulthood, and there are no mechanisms to replace damaged elastin [8,47,58]. This long half-life makes elastin especially vulnerable to cumulative damage in the form of AGEs that can irreversibly crosslink elastin or collagen via nonenzymatic protein glycation [1,36,79]. In addition, elastic arteries are also known to undergo calcification [7,57], as well as enzymatic degradation via MMPs [74,82,83]. Increased collagen deposition, particularly in the collagen-rich adventitial layer, also leads to a stiffer aorta [12]. Oxidative stress [3] and inflammation [11,24] may further influence the structural changes that result in arterial stiffening.

The role of dVSMCs in aortic stiffening has been examined for the most part indirectly in studies of endothelial vasoregulatory dysfunction with age and disease (e.g., reduced bioavailability of nitric oxide) [17,59,62,63,77]. Recently, it has been shown that the stiffness of primary cultured aortic smooth muscle cells is increased in an aging primate model and in a hypertensive rodent model, representing a potential new mechanism for aortic stiffening, but the details of such a mechanism remain unclear [50,56]. Work

from our laboratory employing a multiscale approach has now identified the cortical FA of vascular smooth muscle cells as a key subcellular regulator of aortic stress generation and stiffness [55]. Using a magnetic microneedle to apply piconewton forces to cell-bound, RGD-coated microbeads, we demonstrated that agonist stimulation induces stiffening of the VSMC cortex, and that the small molecule Src-inhibitor PP2 prevents this stiffening. Deconvolution immunofluorescence microscopy revealed that this cortical stiffening parallels agonist-induced increases in FA size, which are also Src dependent. Aorta tissue homogenates probed by Western blot with phosphospecific antibodies indicated increased tyrosine phosphorylation of FA proteins CAS, FAK, and paxillin in response to contractile stimulation, which is indicative of FA remodeling. Both PP2 and FI-14, a small molecule inhibitor of FAK, inhibited agonist-induced increases in tyrosine phosphorylation, as well as agonist-induced contractile force and stiffening, as measured with high-frequency, low-amplitude cyclic stretches that do not break cross-bridges. Taken together with previous studies, these results indicate that agonist stimulation induces actin polymerization, cortical stiffening, and Src-FAK–mediated FA growth and remodeling that strengthens the cortical cytoskeleton and FA to enable tissue stiffening [34,55] (Figure 2). Different FA proteins undergo varying degrees of redistribution in response to agonist stimulation, with proteins like VASP and zyxin that strengthen the link between the FA and cortical actin being most dynamic [48]. This shuffling of FA proteins is linked to actin- and microtubule-dependent endosomal pathways [48], which had previously been implicated in FA recycling in migrating cultured cells [31,54,67], and may help to reinforce high tension-bearing sites.

SUMMARY

The microcirculation, the main regulator of total vascular resistance, plays an important role in cardiovascular disease, but recent studies have indicated a substantial role for the large-diameter elastic aorta as well. Aortic stiffening, a known biomarker for subsequent cardiovascular disease, reduces the impedance mismatch between the aorta and the muscular arteries. As a result, a larger portion of the pulse pressure is transmitted distally to sensitive microvessels in critical organs, including the heart, brain, and kidney. Augmented transmission of the pressure pulse may induce hypertrophy and remodeling of the microcirculation and increase the afterload on the heart.

Clinical measures of aortic stiffness, while noninvasive, incorporate not only the mechanical properties of the vessel wall but also its geometry. It is important to distinguish functional measures of stiffness (PWV, Z_c) from measures of material stiffness (Young's modulus). This will require thorough biomechanical investigations *in vitro* where loading conditions and vessel dimensions can be precisely tracked and controlled.

Recent inquiries into the multifaceted mechanisms of stiffness are forcing investigators to reconsider the aorta as more than simply a passive elastic conduit. Just as dVSMCs are integral to regulating the resistance of the microcirculation, these cells are also integral regulators of aortic stiffness. Further investigations of smooth muscle involvement will not only inform modeling efforts but may also lead to the development of new interventions to mitigate aortic stiffening and reduce the burden of cardiovascular disease.

Thus, the complexity of aortic stiffening with aging and disease warrants a multiscale approach to understand underlying mechanisms and their effects on cardiovascular function. Importantly, the success of this integrative approach will require interdisciplinary collaboration among clinicians, physiologists, biologists, and engineers.

PERSPECTIVE

Aortic stiffness is an early biomarker for cardiovascular disease. The mechanisms of aortic stiffness have long been presumed to be extracellular, although recent studies suggest that vascular smooth muscle cells are important regulators of stiffness. A multiscale approach that integrates insights across fields of study may lead to new therapies for aortic stiffness and cardiovascular disease.

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Abbreviations used:

AGE
advanced glycation end product

CAS
Crk-associated substrate

dVSMC
differentiated (contractile) vascular smooth muscle cell

E
modulus of elasticity (Young's modulus)

FA
focal adhesion

FAK
focal adhesion kinase

MMP
matrix metalloprotease

PWV
pulse wave velocity

Z
impedance

REFERENCES

1. Aronson D Cross-linking of glycated collagen in the pathogenesis of arterial and myocardial stiffening of aging and diabetes. J Hypertens 21: 3–12, 2003. [PubMed: 12544424]
2. Avolio A Input impedance of distributed arterial structures as used in investigations of underlying concepts in arterial haemodynamics. Med Biol Eng Comput 47: 143–151, 2009. [PubMed: 18949501]
3. Bachschmid MM, Schildknecht S, Matsui R, Zee R, Haeussler D, Cohen RA, Pimental D, Loo Bv. Vascular aging: chronic oxidative stress and impairment of redox signaling-consequences for vascular homeostasis and disease. Ann Med 45: 17–36, 2013. [PubMed: 22380696]
4. Bershadsky AD, Balaban NQ, Geiger B. Adhesion-dependent cell mechanosensitivity. Annu Rev Cell Dev Biol 19: 677–695, 2003. [PubMed: 14570586]
5. Bohr DF, Somlyo AP, Sparks HV. Handbook of Physiology. Section 2: The Cardivoascular System. Volume II: Vascular Smooth Muscle, edited by Geiger SR. Baltimore, MD: Oxford University Press, 1980.
6. Cipolla MJ, Gokina NI, Osol G. Pressure-induced actin polymerization in vascular smooth muscle as a mechanism underlying myogenic behavior. FASEB J 16: 72–76,2002. [PubMed: 11772938]
7. Dao HH, Essalihi R, Bouvet C, Moreau P. Evolution and modulation of age-related medial elastocalcinosis: impact on large artery stiffness and isolated systolic hypertension. Cardiovasc Res 66: 307–317, 2005. [PubMed: 15820199]
8. Davis EC. Elastic lamina growth in the developing mouse aorta. J Histochem Cytochem 43: 1115–1123, 1995. [PubMed: 7560894]
9. del Rio A, Perez-Jimenez R, Liu R, Roca-Cusachs P, Fernandez JM, Sheetz MP. Stretching single talin rod molecules activates vinculin binding. Science 323: 638–641, 2009. [PubMed: 19179532]
10. Fincham VJ, Frame MC. The catalytic activity of Src is dispensable for translocation to focal adhesions but controls the turnover of these structures during cell motility. EMBO J 17: 81–92, 1998. [PubMed: 9427743]
11. Fleenor BS, Marshall KD, Rippe C, Seals DR. Replicative aging induces endothelial to mesenchymal transition in human aortic endothelial cells: potential role of inflammation. J Vasc Res 49: 59–64, 2012. [PubMed: 21985896]
12. Fleenor BS, Sindler AL, Marvi NK, Howell KL, Zigler ML, Yoshizawa M, Seals DR. Curcumin ameliorates arterial dysfunction and oxidative stress with aging. Exp Gerontol 48: 269–276, 2013. [PubMed: 23142245]
13. Fung YC. Biomechanics: Circulation, 2nd edn. New York: Springer-Verlag, 1996, 571 pp.
14. Galbraith CG, Yamada KM, Sheetz MP. The relationship between force and focal complex development. J Cell Biol 159: 695–705, 2002. [PubMed: 12446745]
15. Geiger B, Bershadsky A, Pankov R, Yamada KM. Transmembrane extracellular matrix-cytoskeleton crosstalk. Nat Rev Mol Cell Biol 2: 793–805, 2001. [PubMed: 11715046]
16. Geiger T, Zaidel-Bar R. Opening the flood-gates: proteomics and the integrin adhesome. Curr Opin Cell Biol 24: 562–568, 2012. [PubMed: 22728062]
17. Gerhard M, Roddy MA, Creager SJ, Creager MA. Aging progressively impairs endothelium-dependent vasodilation in forearm resistance vessels of humans. Hypertension 27: 849–853, 1996. [PubMed: 8613259]
18. Go AS, Mozaffarian D, Roger VL, Benjamin EJ, Berry JD, Borden WB, Bravata DM, Dai S, Ford ES, Fox CS, Franco S, Fullerton HJ, Gillespie C, Hailpern SM, Heit JA, Howard VJ, Huffman MD, Kissela BM, Kittner SJ, Lackland DT, Lichtman JH, Lisabeth LD, Magid D, Marcus GM, Marelli A, Matchar DB, McGuire DK, Mohler ER, Moy CS, Mussolino ME, Nichol G, Paynter NP, Schreiner PJ, Sorlie PD, Stein J, Turan TN, Virani SS, Wong ND, Woo D, Turner MB; American Heart Association Statistics Committee and Stroke Statistics Subcommittee. Heart disease and stroke statistics–2013 update: a report from the American Heart Association. Circulation 127: e6–e245, 2013. [PubMed: 23239837]
19. Greenwald SE. Ageing of the conduit arteries. J Pathol 211: 157–172, 2007. [PubMed: 17200940]

20. Gunst SJ, Zhang WW. Actin cytoskeletal dynamics in smooth muscle: a new paradigm for the regulation of smooth muscle contraction. Am J Physiol Cell Physiol 295: C576–C587, 2008. [PubMed: 18596210]
21. Guo X, Kassab GS. Variation of mechanical properties along the length of the aorta in C57bl/6 mice. Am J Physiol Heart Circ Physiol 285: H2614–H2622, 2003. [PubMed: 14613915]
22. Guo X, Kono Y, Mattrey R, Kassab GS. Morphometry and strain distribution of the C57BL/6 mouse aorta. Am J Physiol Heart Circ Physiol 283: H1829–H1837, 2002. [PubMed: 12384460]
23. Guyton AC, Hall JE. Textbook of Medical Physiology, 11th edn. Philadelphia: Elsevier Saunders, 2006, xxxv, 1116 pp.
24. Guzik TJ, Hoch NE, Brown KA, McCann LA, Rahman A, Dikalov S, Goronzy J, Weyand C, Harrison DG. Role of the T cell in the genesis of angiotensin II induced hypertension and vascular dysfunction. J Exp Med 204: 2449–2460, 2007. [PubMed: 17875676]
25. Hanon O, Haulon S, Lenoir H, Seux ML, Rigaud AS, Safar M, Girerd X, Forette F. Relationship between arterial stiffness and cognitive function in elderly subjects with complaints of memory loss. Stroke, 36: 2193–2197, 2005. [PubMed: 16151027]
26. Holzapfel G Chapter 11. Collagen in arterial walls: biomechanical aspects. In: Collagen. Structure and Mechanics, edited by Fratzl P New York: Springer, 2008, pp. 285–324.
27. Humphrey JD. Vascular adaptation and mechanical homeostasis at tissue, cellular, and sub-cellular levels. Cell Biochem Biophys 50: 53–78, 2008. [PubMed: 18209957]
28. Hynes RO. Changing partners. Science 300: 755–756, 2003. [PubMed: 12730590]
29. Ingber DE. Mechanobiology and diseases of mechanotransduction. Ann Med 35: 564–577, 2003. [PubMed: 14708967]
30. Kaess BM, Rong J, Larson MG, Hamburg NM, Vita JA, Levy D, Benjamin EJ, Vasan RS, Mitchell GF. Aortic stiffness, blood pressure progression, and incident hypertension. JAMA 308: 875–881, 2012. [PubMed: 22948697]
31. Kaplan KB, Swedlow JR, Varmus HE, Morgan DO. Association of p60c-src with endosomal membranes in mammalian fibroblasts. J Cell Biol 118: 321–333, 1992. [PubMed: 1378446]
32. Kassab GS. Biomechanics of the cardiovascular system: the aorta as an illustratory example. J R Soc Interface 3: 719–740, 2006. [PubMed: 17015300]
33. Kim HR, Appel S, Vetterkind S, Gangopadhyay SS, Morgan KG. Smooth muscle signalling pathways in health and disease. J Cell Mol Med 12: 2165–2180, 2008. [PubMed: 19120701]
34. Kim HR, Gallant C, Leavis PC, Gunst SJ, Morgan KG. Cytoskeletal remodeling in differentiated vascular smooth muscle is actin isoform dependent and stimulus dependent. Am J Physiol Cell Physiol 295: C768–C778, 2008. [PubMed: 18596213]
35. Klein R, Klein BE, Tomany SC, Cruickshanks KJ. The association of cardiovascular disease with the long-term incidence of age-related maculopathy: the Beaver Dam Eye Study. Ophthalmology 110: 1273–1280, 2003. [PubMed: 12799274]
36. Konova E, Baydanoff S, Atanasova M, Velkova A. Age-related changes in the glycation of human aortic elastin. Exp Gerontol 39: 249–254, 2004. [PubMed: 15036419]
37. Martinez-Lemus LA, Wu X, Wilson E, Hill MA, Davis GE, Davis MJ, Meininger GA. Integrins as unique receptors for vascular control. J Vasc Res 40: 211–233, 2003. [PubMed: 12902635]
38. Mathers CD, Loncar D. Projections of global mortality and burden of disease from 2002 to 2030. PLoS Med 3: e442, 2006. [PubMed: 17132052]
39. Matsumoto T, Nagayama K. Tensile properties of vascular smooth muscle cells: bridging vascular and cellular biomechanics. J Biomech 45: 745–755, 2012. [PubMed: 22177671]
40. Mitchell GF. Clinical achievements of impedance analysis. Med Biol Eng Comput 47: 153–163, 2009. [PubMed: 18853214]
41. Mitchell GF, Tardif JC, Arnold JM, Marchiori G, O'Brien TX, Dunlap ME, Pfeffer MA. Pulsatile hemodynamics in congestive heart failure. Hypertension 38: 1433–1439, 2001. [PubMed: 11751731]
42. Mitra SK, Hanson DA, Schlaepfer DD. Focal adhesion kinase: in command and control of cell motility. Nat Rev Mol Cell Biol 6: 56–68, 2005. [PubMed: 15688067]

43. Morishita R, Gibbons GH, Ellison KE, Lee W, Zhang L, Yu H, Kaneda Y, Ogihara T, Dzau VJ. Evidence for direct local effect of angiotensin in vascular hypertrophy. In vivo gene transfer of angiotensin converting enzyme. J Clin Invest 94: 978–984, 1994. [PubMed: 8083382]
44. Nichols WW, O'Rourke MF, Hartley C. McDonald's Blood Flow in Arteries: Theoretical, Experimental and Clinical Principles, Vol. 340809418. London: Hodder Arnold, 2005.
45. Opazo Saez A, Zhang W, Wu Y, Turner CE, Tang DD, Gunst SJ. Tension development during contractile stimulation of smooth muscle requires recruitment of paxillin and vinculin to the membrane. Am J Physiol Cell Physiol 286: C433–C447, 2004. [PubMed: 14576084]
46. Parker KH. A brief history of arterial wave mechanics. Med Biol Eng Comput 47: 111–118, 2009. [PubMed: 19198914]
47. Powell JT, Vine N, Crossman M. On the accumulation of D-aspartate in elastin and other proteins of the ageing aorta. Atherosclerosis 97: 201–208, 1992. [PubMed: 1466664]
48. Poythress RH, Gallant C, Vetterkind S, Morgan KG. Vasoconstrictor-induced endocytic recycling regulates focal adhesion protein localization and function in vascular smooth muscle. Am J Physiol Cell Physiol 305: C215–C227, 2013. [PubMed: 23703522]
49. Puklin-Faucher E, Sheetz MP. The mechanical integrin cycle. J Cell Sci 122: 179–186, 2009. [PubMed: 19118210]
50. Qiu H, Zhu Y, Sun Z, Trzeciakowski JP, Gansner M, Depre C, Resuello RR, Natividad FF, Hunter WC, Genin GM, Elson EL, Vatner DE, Meininger GA, Vatner SF. Short communication: vascular smooth muscle cell stiffness as a mechanism for increased aortic stiffness with aging. Circ Res 107: 615–619, 2010. [PubMed: 20634486]
51. Rachev A, Hayashi K. Theoretical study of the effects of vascular smooth muscle contraction on strain and stress distributions in arteries. Ann Biomed Eng 27: 459–468, 1999. [PubMed: 10468230]
52. Roach MR, Burton AC. The reason for the shape of the distensibility curves of arteries. Can J Biochem Physiol 35: 681–690, 1957. [PubMed: 13460788]
53. Roca-Cusachs P, Iskratsch T, Sheetz MP. Finding the weakest link: exploring integrin-mediated mechanical molecular pathways. J Cell Sci 125: 3025–3038, 2012. [PubMed: 22797926]
54. Sandilands E, Cans C, Fincham VJ, Brunton VG, Mellor H, Prendergast GC, Norman JC, Superti-Furga G, Frame MC. RhoB and actin polymerization coordinate Src activation with endosome-mediated delivery to the membrane. Dev Cell 7: 855–869, 2004. [PubMed: 15572128]
55. Saphirstein RJ, Gao YZ, Jensen MH, Gallant CM, Vetterkind S, Moore JR, Morgan KG. The focal adhesion: a regulated component of aortic stiffness. PLoS One 8: e62461, 2013. [PubMed: 23626821]
56. Sehgel NL, Zhu Y, Sun Z, Trzeciakowski JP, Hong Z, Hunter WC, Vatner DE, Meininger GA, Vatner SF. Increased vascular smooth muscle cell stiffness; a novel mechanism for aortic stiffness in hypertension. Am J Physiol Heart Circ Physiol 305: H1281–H1287, 2013. [PubMed: 23709594]
57. Shao JS, Cai J, Towler DA. Molecular mechanisms of vascular calcification: lessons learned from the aorta. Arterioscler Thromb Vasc Biol 26: 1423–1430, 2006. [PubMed: 16601233]
58. Shapiro SD, Endicott SK, Province MA, Pierce JA, Campbell EJ. Marked longevity of human lung parenchymal elastic fibers deduced from prevalence of D-aspartate and nuclear weapons-related radiocarbon. J Clin Invest 87: 1828–1834, 1991. [PubMed: 2022748]
59. Sindler AL, Fleenor BS, Calvert JW, Marshall KD, Zigler ML, Lefer DJ, Seals DR. Nitrite supplementation reverses vascular endothelial dysfunction and large elastic artery stiffness with aging. Aging Cell 10: 429–437, 2011. [PubMed: 21276184]
60. Small JV, Gimona M. The cytoskeleton of the vertebrate smooth muscle cell. Acta Physiol Scand 164: 341–348, 1998. [PubMed: 9887957]
61. Somlyo AV. Ultrastructure of vascular smooth muscle. In: Handbook of Physiology. The Cardiovascular System. Vascular Smooth Muscle, edited by Soc AP, Bethesda MD: A.P. Soc, 1980, p. 33–67.
62. Soucy KG, Ryoo S, Benjo A, Lim HK, Gupta G, Sohi JS, Elser J, Aon MA, Nyhan D, Shoukas AA, Berkowitz DE. Impaired shear stress-induced nitric oxide production through decreased NOS phosphorylation contributes to age-related vascular stiffness. J Appl Physiol 101: 1751–1759, 2006. [PubMed: 17106067]

63. Stewart AD, Millasseau SC, Kearney MT, Ritter JM, Chowienczyk PJ. Effects of inhibition of basal nitric oxide synthesis on carotid-femoral pulse wave velocity and augmentation index in humans. Hypertension 42: 915–918, 2003. [PubMed: 12975386]
64. Tang DD, Wu MF, Opazo Saez AM, Gunst SJ. The focal adhesion protein paxillin regulates contraction in canine tracheal smooth muscle. J Physiol 542: 501–513, 2002. [PubMed: 12122148]
65. Taylor MG. An approach to an analysis of the arterial pulse wave. I. Oscillations in an attenuating line. Phys Med Biol 1: 258–269, 1957. [PubMed: 13419530]
66. Taylor MG. An approach to an analysis of the arterial pulse wave. II. Fluid oscillations in an elastic pipe. Phys Med Biol 1: 321–329, 1957. [PubMed: 13452838]
67. Tu C, Ortega-Cava CF, Winograd P, Stanton MJ, Reddi AL, Dodge I, Arya R, Dimri M, Clubb RJ, Naramura M, Wagner KU, Band V, Band H. Endosomal-sorting complexes required for transport (ESCRT) pathway-dependent endosomal traffic regulates the localization of active Src at focal adhesions. Proc Natl Acad Sci U S A 107: 16107–16112, 2010. [PubMed: 20805499]
68. Vande Geest JP, Sacks MS, Vorp DA. Age dependency of the biaxial biomechanical behavior of human abdominal aorta. J Biomech Eng 126: 815–822, 2004. [PubMed: 15796340]
69. Verhave JC, Fesler P, du Cailar G, Ribstein J, Safar ME, Mimran A. Elevated pulse pressure is associated with low renal function in elderly patients with isolated systolic hypertension. Hypertension 45: 586–591, 2005. [PubMed: 15738348]
70. Vito RP, Dixon SA. Blood vessel constitutive models-1995–2002. Annu Rev Biomed Eng 5: 413–439, 2003. [PubMed: 12730083]
71. Wagenseil JE, Mecham RP. Vascular extracellular matrix and arterial mechanics. Physiol Rev 89: 957–989, 2009. [PubMed: 19584318]
72. Wagenseil JE, Nerurkar NL, Knutsen RH, Okamoto RJ, Li DY, Mecham RP. Effects of elastin haploinsufficiency on the mechanical behavior of mouse arteries. Am J Physiol Heart Circ Physiol 289: H1209–H1217, 2005. [PubMed: 15863465]
73. Waldstein SR, Rice SC, Thayer JF, Najjar SS, Scuteri A, Zonderman AB. Pulse pressure and pulse wave velocity are related to cognitive decline in the Baltimore Longitudinal Study of Aging. Hypertension 51: 99–104, 2008. [PubMed: 18025297]
74. Wang M, Lakatta EG. Altered regulation of matrix metalloproteinase-2 in aortic remodeling during aging. Hypertension 39: 865–873, 2002. [PubMed: 11967241]
75. Webb DJ, Donais K, Whitmore LA, Thomas SM, Turner CE, Parsons JT, Horwitz AF. FAK-Src signalling through paxillin, ERK and MLCK regulates adhesion disassembly. Nat Cell Biol 6: 154–161, 2004. [PubMed: 14743221]
76. Westerhof N, Lankhaar JW, Westerhof BE. The arterial Windkessel. Med Biol Eng Comput 47: 131–141, 2009. [PubMed: 18543011]
77. Wilkinson IB, Franklin SS, Cockcroft JR. Nitric oxide and the regulation of large artery stiffness: from physiology to pharmacology. Hypertension 44: 112–116, 2004. [PubMed: 15262901]
78. Wolfenson H, Henis YI, Geiger B, Bershadsky AD. The heel and toe of the cell's foot: a multifaceted approach for understanding the structure and dynamics of focal adhesions. Cell Motil Cytoskelet 66: 1017–1029, 2009.
79. Wolffenbuttel BH, Boulanger CM, Crijns FR, Huijberts MS, Poitevin P, Swennen GN, Vasan S, Egan JJ, Ulrich P, Cerami A, Lévy BI. Breakers of advanced glycation end products restore large artery properties in experimental diabetes. Proc Natl Acad Sci U S A 95: 4630–4634, 1998. [PubMed: 9539789]
80. Wolinsky H, Glagov S. Structural basis for the static mechanical properties of the aortic media. Circ Res 14: 400–413, 1964. [PubMed: 14156860]
81. World Health Organization. Global Status Report on Noncommunicable Diseases 2010. Geneva, Switzerland: World Health Organization, 2011, ix, 162 pp.
82. Yasmin, McEniery CM, O'Shaughnessy KM, Harnett P, Arshad A, Wallace S, Maki-Petaja K, McDonnell B, Ashby MJ, Brown J, Cockcroft JR, Wilkinson IB. Variation in the human matrix metalloproteinase-9 gene is associated with arterial stiffness in healthy individuals. Arterioscler Thromb Vasc Biol 26: 1799–1805, 2006. [PubMed: 16709939]
83. Yasmin, McEniery CM, Wallace S, Dakham Z, Pulsalkar P, Maki-Petaja K, Ashby MJ, Cockcroft JR, Wilkinson IB. Matrix metalloproteinase-9 (MMP-9), MMP-2, and serum elastase activity are

associated with systolic hypertension and arterial stiffness. Arterioscler Thromb Vasc Biol 25: 372, 2005. [PubMed: 15556929]

84. Zaidel-Bar R, Ballestrem C, Kam Z, Geiger B. Early molecular events in the assembly of matrix adhesions at the leading edge of migrating cells. J Cell Sci 116: 4605–4613, 2003. [PubMed: 14576354]
85. Zaidel-Bar R, Cohen M, Addadi L, Geiger B. Hierarchical assembly of cell-matrix adhesion complexes. Biochem Soc Trans 32: 416–420, 2004. [PubMed: 15157150]
86. Zaidel-Bar R, Itzkovitz S, Ma'ayan A, Iyengar R, Geiger B. Functional atlas of the integrin adhesome. Nat Cell Biol 9: 858–867, 2007. [PubMed: 17671451]
87. Zhang W, Gunst SJ. Interactions of airway smooth muscle cells with their tissue matrix: implications for contraction. Proc Am Thorac Soc 5: 32–39, 2008. [PubMed: 18094082]
88. Zhang X, Jiang G, Cai Y, Monkley SJ, Critchley DR, Sheetz MP. Talin depletion reveals independence of initial cell spreading from integrin activation and traction. Nat Cell Biol 10: 1062–1068, 2008. [PubMed: 19160486]

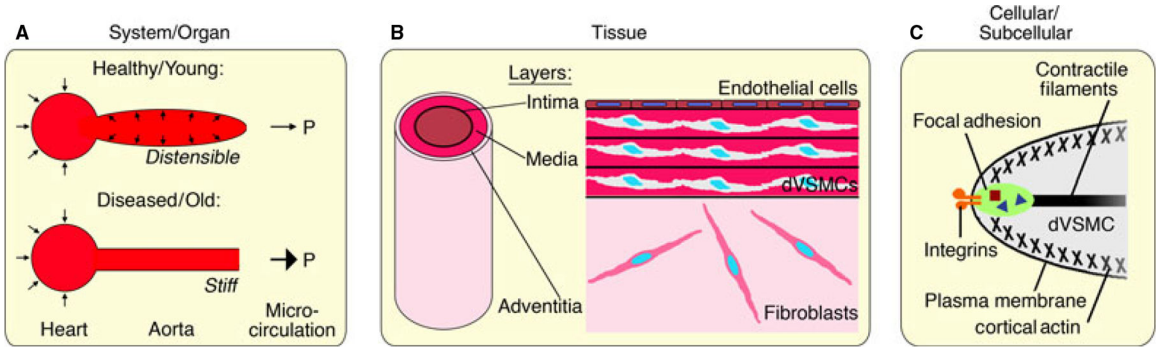

Figure 1.

The aorta across length scales. An integrative consideration of structure and function at different biological levels of scale is necessary to understand cardiovascular physiology and biomechanics in health and disease. (**A**) System/organ level. Top: Contraction of the heart ejects blood into the arterial systemic circulation. The blood first enters the aorta, which distends and then recoils to propel the blood downstream. Bottom: With an aging- or disease-related increase in aortic stiffness, the aorta is less distensible and absorbs less of the energy of the pressure pulse. (**B**) Tissue level. Aortic stiffness dictates aortic function, and aortic stiffness is determined by the structural organization and composition of the vessel wall. The wall is arranged in three main layers (intima, media, and adventitia), each containing a characteristic cell type (endothelial cells, vascular smooth muscle cells, and fibroblasts, respectively). (**C**) Cellular/subcellular level. Vascular stiffness depends critically on both the extracellular matrix and the smooth muscle cells embedded in the matrix. VSMCs are physically connected to their surrounding matrix by transmembrane integrin receptors, which are connected via the actin cytoskeleton to contractile filaments responsible for force production and regulation of stiffness.

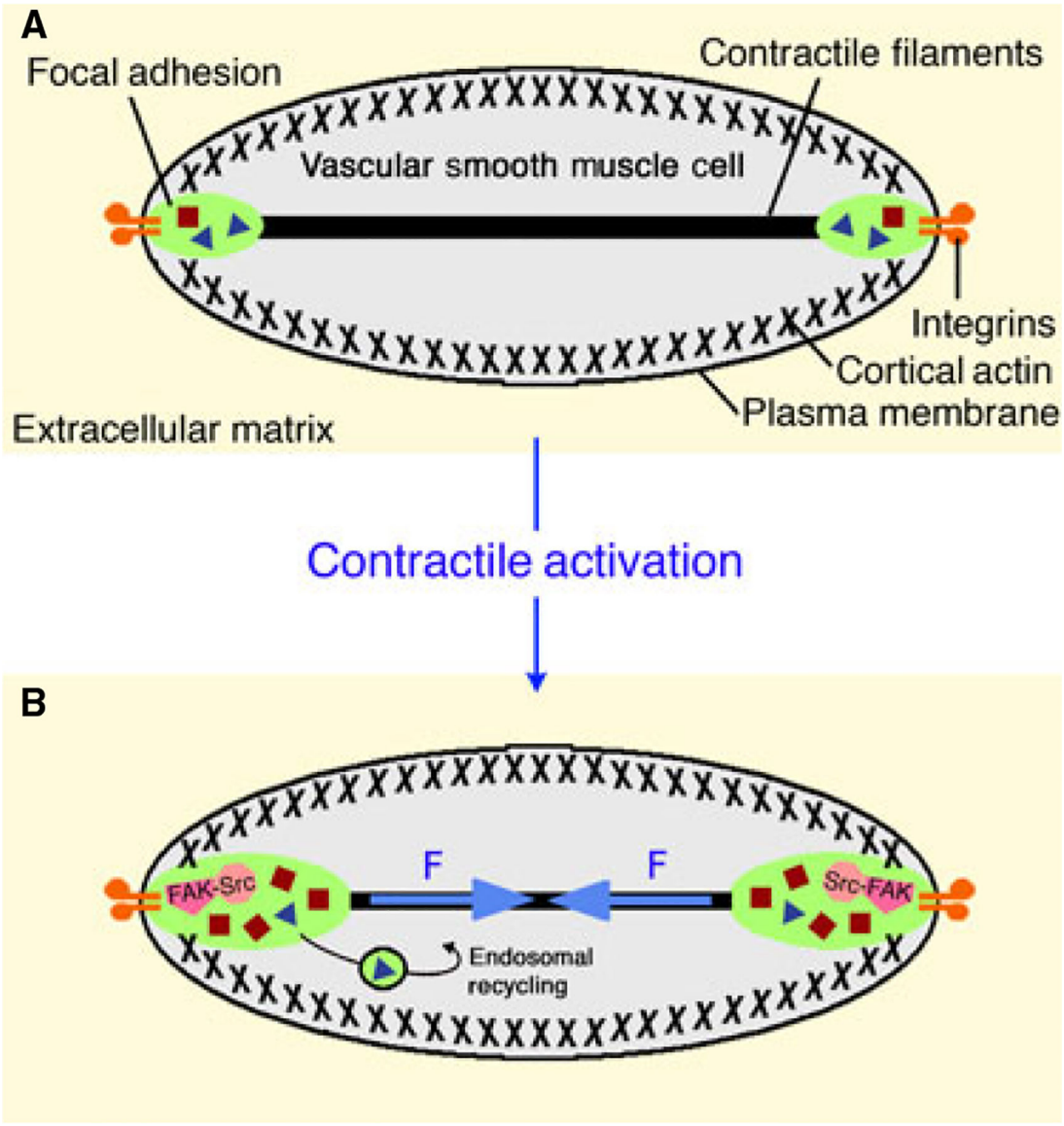

Figure 2.

Model of FA regulation of aortic stiffness. Agonist- and tension-induced growth and remodeling of FA in aortic VSMCs strengthen cell–matrix adhesion to facilitate force transmission and stiffening of the aorta. Differential redistribution of FA proteins is facilitated by endosomal recycling pathways. Modified from [55].