Published in final edited form as:
Circulation. 1997 January 21; 95(2): 522–528.

Coronary Microcirculation in Health and Disease Summary of an NHLBI Workshop

William M. Chilian, PhD with contributions from the NHLBI Workshop Participants*

Abstract

This article summarizes a 2-day workshop on the coronary microcirculation held in Bethesda, Md, in September 1994 and sponsored by the National Heart, Lung, and Blood Institute of the National Institutes of Health. The workshop explored a variety of topics pertaining to coronary microvascular physiology and pathophysiology. The latest methodologies that are being used to investigate the coronary microvasculature, including endoscopic microscopy of the intramural coronary microvasculature and micro–x-ray computerized tomography, were discussed. The most recent advances in the regulation of the coronary microcirculation—for example, myogenic and flow-dependent responses, K_{ATP} channels, and regional heterogeneity—were reported. The workshop touched on the relation of the microcirculation to clinically important conditions and offered recommendations for future research in this important area. Comparisons are made to recent advances in the peripheral circulation and current gaps in our knowledge concerning the coronary microcirculation. In recent years, research on the coronary microcirculation has made substantial advances, in part as a result of investigations in the peripheral microcirculation but also because of the application of unique methodologies. This research is providing new ways to investigate abnormalities of myocardial perfusion, an area of inquiry that until recently has been limited to examination of coronary pressure-flow relationships.

Keywords

microcirculation; endothelium-derived factors; syndrome X; ischemia; myocardium

This article represents a summary of a 2-day workshop on the coronary microcirculation held in Bethesda, Md, in September 1994 and sponsored by the National Heart, Lung, and Blood Institute of the National Institutes of Health. The workshop explored the characteristics of the coronary microcirculation that distinguish it from other microcirculatory beds, discussed the latest methodology being used to investigate this vascular bed, touched on its relation to clinically important conditions, and offered recommendations for future research in this important area. In recent years, our understanding of the coronary microcirculation has advanced substantially, in part as a result of investigations in the peripheral microcirculation but also because of the application of unique methodologies. This research is providing new ways to investigate abnormalities of

Correspondence to Alfred Small, PhD, Vascular Research Program, Division of Heart and Vascular Diseases, National Heart, Lung, and Blood Institute, Two Rockledge Center, 6701 Rockledge Dr, MSC 7956, Bethesda, MD 20892-7956.

*Participants in the NHLBI Workshop are listed in the "Appendix."

myocardial perfusion, an area of inquiry that until recently has been limited to examination of coronary pressure-flow relationships.

This article is organized into five topic areas: (1) recent methods that enable new measurements of coronary microvascular function, (2) heterogeneity of the coronary microcirculation, (3) recent physiological findings, (4) clinical aspects of the coronary microcirculation, and (5) future directions. Within each of these areas, comparisons are made to recent advances in the peripheral circulation, and gaps in our knowledge about the coronary microcirculation are identified.

Methodology and Measurements in the Coronary Microcirculation

The investigation of coronary microcirculatory function entails several difficulties, notably gross movement attributed to cardiac contraction, that are not found in other microcirculatory beds. For this reason, the development of unique methodological approaches for visualizing and manipulating these vessels has significantly advanced the field. Microvessels on the epicardial surface of the beating heart have been studied since about 1980 with a microscope-video system that freezes motion of the vessel through the use of stroboscopic epi-illumination synchronized with the heart cycle.1 Drawbacks of the method are that the measurements are restricted to the subepicardial microcirculation, which most likely has attributes that render it different from the microvasculatures from other ventricular regions, and that the procedure is invasive.

Recently, a method for directly viewing subendocardial and intramural vessels in the beating heart was developed.2,3 This technique uses a video microscope with a needle probe (image-transferring conduit) that serves to project an image over a distance of several centimeters to the objective lens. The needle probe can be placed near the endocardial surface in a protected blood-free space created by a dough-nut-shaped balloon, or it can be inserted directly into the myocardium. This procedure, too, is invasive. The temporal and spatial limits of this methodology are being improved by high-definition and high-speed video systems, and ultimately, measurements of flow should be feasible. This technique is most valuable in assessing the impact of various regulatory mechanisms on subepicardial and subendocardial vascular diameters, and it opens investigations into areas of the coronary microcirculation that were previously unapproachable.4,5

The only current experimental methodology that has the potential to noninvasively provide images of the coronary microcirculation is x-ray microcomputerized tomography (micro-CT).6 Micro-CT imaging allows three-dimensional views of the entire coronary microcirculation and provides a useful means of studying vascular structure and function.7 For example, when the coronary vascular tree of a rat is injected with a polymerized contrast material, one can visualize the entire tree in three dimensions. Micro-CT can scan 1-mL volumes at resolutions down to ≈10 μm.3,8 The potential for imaging the intramural vasculature by micro-CT is enormous. Not only can detailed analyses of coronary morphometry in intact tissues be evaluated in three dimensions, but noninvasive studies of regulation of the intramural coronary microcirculation also are theoretically possible. This

technique, however, is still in its infancy, and measurements have been obtained only from arrested preparations.

In vitro methodologies also have been used to study the coronary microcirculation. These strategies have been applied as a way to avoid the limitations imposed by the motion and thickness of the intact heart. Microvessels of interest can be dissected from any region of the heart, including the human heart, and can be cannulated and perfused at varied pressures and flows.9–11 Isolated vessels have been used to measure many vasomotor responses of coronary arterioles and muscular venules and the regulation of permeability in isolated venules.12 Results of investigations using this technique to measure vasoactive responses will be described later in this article. In addition, technological advances in cell isolation and identification involving fluorescence cell sorting, magnetic beads, and antibodies have greatly facilitated the feasibility of isolating endothelial cells from specific microvascular beds.13–15 The newest methods offer the promise of isolating endothelial cells derived from specific segments of the microcirculation with high yields without requiring extensive dissection.

Heterogeneity in the Coronary Microcirculation

Heterogeneity of pathophysiological changes in the heart is evidenced by the well-known vulnerability of the left subendocardium to ischemia, and it is of great clinical importance not only in coronary heart disease but also in the case of subendocardial ischemia seen in many congenital or acquired heart diseases in which the coronary arteries are normal.16 Spatial variability of flow may account for the greater susceptibility of the subendocardium to various vascular and cardiac pathological conditions. Experimental observations suggest that the distribution of regional flows in the heart is not random but rather exhibits a pattern: regional flows are more similar to those of near-neighbor regions than they are to more distant locations.17–20 The degree of near-neighbor correlation and its diminution with distance are independent of the size of the volume elements in which the flows are averaged, a pattern of self-similarity shown by fractal structures. Studies using artificial networks of vessels based on the known pattern of vessel branching indicate that this self-similarity arises from the dichotomous branching of vessels according to simple recursion rules.21,22

Heterogeneity of flow has important implications for the measurement of flows in the myocardium. The response of the whole heart to a submaximal perturbation (such as hypoxia) can be misleading, because it will include both responding and nonresponding regions; reliable correlations can be drawn only from the use of sufficiently small tissue samples.23 Clinically, this represents a problem for defining an adequate average flow, eg, determining the adequacy of coronary flow in a patient from a spatially averaged sample. The cause of spatial variations in heterogeneity of myocardial perfusion has not yet been completely resolved, but it appears that spatial variations can be attributed to regional variations in metabolism. This suggestion was made from results indicating that regional flow and transport capacity are closely related.24 Heterogeneity is also evident in the structural, biochemical, and functional features of individual endothelial cells and is, to a great extent, determined by the organ of origin and by the vascular level (artery, arteriole, venule, capillary) at which they occur. Thus, understanding the mechanisms responsible for

heterogeneity is imperative for complete comprehension of coronary microvascular structure and function.25

Recent Physiological Findings

Anatomically and functionally distinct categories of myocardial vessels segregated into discrete areas have been called vascular microdomains because they are reminiscent of the protein microdomains that provide for specialization of functions. For example, water and solutes are exchanged primarily in capillaries and postcapillary (pericytic) venules, whereas coronary vascular resistance resides predominantly in arterioles. These functional domains can be further distinguished by the regulatory mechanisms that dominate a specific function. For instance, neurogenic, metabolic, myogenic, and shear stress–induced mechanisms dominate resistance at specific microvascular sites.26 The organization of vascular microdomains appears to be influenced by factors that induce phenotypic specialization of endothelial and smooth muscle cells. For example, the expression of α_1- and α_2-adrenergic receptors, which differs among various vascular segments of the intact microcirculation, 27,28 is modulated by stretch in cultured coronary vascular smooth muscle.29 It is worth emphasizing that the causal factors for this organizational scheme in the intact coronary vasculature have not been unequivocally identified.

Traditionally, the close matching of coronary blood flow to myocardial oxygen consumption has been attributed to metabolic mechanisms, although the precise mediators have eluded discovery.30 Recent studies indicate that myogenic and endothelial mechanisms strongly influence diameters of coronary microvessels through the transduction of intravascular pressure (stretch) and flow (shear stress). Myogenic responsiveness in the coronary microcirculation was demonstrated directly in 1988 and was shown to involve vessels of a size that contributes significantly to coronary microvascular resistance in vivo.10 Myogenic dilatation and constriction are potentially autoregulatory, and a fall in arteriolar pressure during either coronary occlusion or metabolic vasodilatation would be expected to reduce coronary vascular resistance through this mechanism. Although it is difficult to invoke flow-dependent dilation in autoregulation, this mechanism may potentially affect the network response to metabolic vasodilation (discussed below).9,31 Flow-induced dilatation was also demonstrated in isolated coronary venules; this behavior may be important during arteriolar dilatation, when the venous contribution to total coronary resistance increases from about 10% to 30%.32 In this situation, flow-dependent dilatation of coronary venules would limit the rise in capillary pressure that occurs when arteriolar resistance decreases and thus would help prevent excessive trans-capillary filtration and the possibility of myocardial edema.

Longitudinal gradients for pressure- and flow-dependent responses can be integrated into a hypothetical system that could match coronary blood flow to myocardial metabolic demands.26,33 Coronary arterioles with diameters between 120 and 150 μm are the segments most sensitive to flow, whereas small arterioles (30 to 60 μm) seem to exhibit the greatest myogenic responsiveness, and arterioles <30 μm appear to be the most sensitive to metabolic stimuli.26 According to this scheme, the smallest arterioles dilate during increased metabolic demand, lowering microvascular resistance so that myocardial perfusion increases. As the upstream arteriolar pressure falls, myogenic dilatation of slightly

larger arterioles and thus a further decrease in resistance occur. Increased flow in larger arterioles upstream stimulates flow-dependent dilatation, further reducing network resistance. Flow-dependent dilatation of upstream arterioles also would transmit increased pressure and flow to downstream arterioles so that the latter retain tone and thus their sensitivity to further changes in metabolic demand, although on the basis of recent observations, this scheme remains hypothetical, because there is still no information about the role of myogenic and flow-dependent regulation of arteriolar tone in the intact heart. Even metabolic vasodilation, a seminal mechanism involved in the balance between oxygen supply and demand, remains poorly understood. Elucidating the contributions of these microvascular mechanisms to metabolic hyperemia is imperative for understanding coronary vascular physiology.

In the peripheral circulation, communication of vasoactive responses via cell-cell communication has been widely acknowledged for several years. These mechanisms, although not yet identified in coronary microvessels, may participate in the coordination of coronary vasomotor responses. Electrical communication between cells in arterioles was suggested from measurements of intracellular potentials, which revealed that resting membrane potentials are virtually identical in smooth muscle and endothelium.34 Transient changes in membrane potential induced with agonists were identical in smooth muscle and endothelial cells, and the electrical signal traveled with a similar conduction velocity in both types of cells. These results suggest that in the arteriolar wall, endothelial and vascular smooth muscle cells are united into an electrical syncytium. This hypothesis was strengthened by the demonstration that antibodies to connexin 43 and connexin 40 revealed pericellular plaques of both gap junctional proteins35 and that gap junctional tracers moved bidirectionally between adjacent smooth muscle or between adjacent endothelial cells.36–37 A surprising finding was that tracers moved from endothelial cells to the overlying smooth muscle but not in the reverse direction.38 This system has the potential to unite endothelial and smooth muscle cells, thus converting the actions of discrete cellular elements into a functional microvessel, and could provide for longitudinal coordination of the vasculature. Again, it should be emphasized that, to date, there have been no such studies of propagated responses or intercellular communication in the coronary microcirculation. Therefore, the importance of these modes of network communication for coronary vasomotor control is unresolved.

Relative to our knowledge of the vasomotor control in the coronary microcirculation of experimental models, there is a paucity of knowledge concerning such regulation in human coronary microvessels. In addition, there are indications of important species differences in the behavior of the myocardial microcirculation. For example, in human coronary vessels, acetylcholine shows pharmacological asymmetry; it is a dilator when administered intraluminally but a constrictor when applied adventitially.39–40 This asymmetry is not shown by other endothelium-dependent dilators, such as histamine and bradykinin.39,40 Another unique characteristic of human coronary artery contractile responses is the development of endothelium-independent oscillatory contractions in both epicardial conduit and intramyocardial resistance arteries.41 Studies showing that these oscillatory contractions can be abolished with ouabain, inhibition of the slow, voltage-dependent calcium channels with nifedipine, or activation of nitric oxide production with histamine and thrombin provide

only limited clues to the mechanisms responsible for this phenomenon. Attempts to associate this behavior with pathological processes have also met with little success; studies so far have failed to show a link between the oscillations and presence of free fatty acids, cholesterol, leukotriene metabolites, lipid accumulation, or the presence of inflammatory cells.

The preceding discussion addresses primarily the adaptation of coronary resistance vessels to acute physiological stresses, such as an increase in myocardial metabolism. Yet it is important to emphasize that the coronary micro-circulation also appears to adapt to chronic physiological stresses. Exercise appears to induce numerous adaptive changes in the coronary circulation that may be responsible, in part, for the reduction in coronary heart disease that is attributed to exercise training.42 Blood flow capacity and transvascular exchange capacity are increased by exercise training, possibly because of remodeling of the coronary vascular bed and as a result of alterations in vascular control.43 Although it is known that exercise training results in growth of proximal coronary arteries, little is known about remodeling in the microcirculation.42,44 Exercise-trained pigs have increased numbers and lengths of arterioles, but training does not appear to alter coronary capillarity. 42,44 Exercise training may also alter vascular control processes in the coronary microcirculation.42,45,46 Microvascular responses to mechanical stimuli such as stretch and flow also appear to be altered by exercise training,45,46 and training may increase the amount of nitric oxide synthase in the coronary microvasculature.47 There is now preliminary evidence that exercise training alters the regulation of coronary vascular permeability.

In the coronary circulation, the regulation of permeability is relatively unexplored. Only a few studies have examined this quantitatively (in isolated venules), in contrast to the peripheral circulation, on which there is a large body of literature.12,48–50 When endothelial cells of mesenteric microvessels are exposed to inflammatory agents, there appears to be an association between intracellular calcium and permeability.51 Most of the initial increase in intracellular calcium is due to influx from a passive conductance channel, although a portion of the intracellular increase is due to release from internal stores.52,53 Thus, hyperpolarization of the endothelial cell membrane potentiates the initial increase in intracellular calcium and microvessel permeability, whereas depolarization has the opposite effect.54 Elevated levels of intracellular calcium do not directly increase permeability: a cAMP analogue attenuated the increase in permeability caused by ATP but did not affect calcium kinetics.55 Two of the fundamental unanswered questions about the regulation of permeability in the heart are, What are the primary regulatory mechanisms, and Can alterations in exchange vessels contribute to the manifestations of various cardiac pathologies? Further observations are needed to gain insight into which calcium-dependent and -independent enzymes modulate structures that determine microvessel permeability. Targets of regulation might include the proteins forming the tight junctional complex and the adherens region of the intercellular junction, the proteins regulating integrin–extracellular matrix interactions, proteins of the endothelial cell glycocalyx, and factors that modulate the production of nitric oxide.

Another important factor in the regulation of vascular permeability in all microcirculatory beds is the production of reactive oxygen species by adherent neutrophils.56,57 This is considered an especially significant determinant of events produced by reperfusion after ischemia, both to vascular function and to the well-being of the underlying tissue. An important area of research relates to the mechanisms of neutrophil-induced changes in vascular permeability. Are the increase in permeability and the breakdown of the endothelial barrier related to the production of free radicals? Secretion of proteases? Production of nitric oxide? The process of diapedesis? Furthermore, even the extent to which neutrophils contribute to myocardial reperfusion injury is controversial.58–60 These and many other questions must be answered to more fully understand the pathophysiological sequelae of reperfusion injury in the heart.

Clinical Importance of the Coronary Microvascular Pathologies

Although acute and chronic ischemic syndromes are commonly due to coronary flow–limiting atherosclerotic plaques in epicardial coronary arteries, ≈10% to 20% of patients undergoing cardiac catheterization are found to have normal coronary angiograms.61 In some, cardiomyopathies such as familial hypertrophic cardiomyopathy and hypertensive left ventricular hypertrophy produce morphological changes in the intramyocardial small arteries that may limit appropriate flow delivery to the myocardium during stress, resulting in ischemia.62,63 However, in the majority of patients with angina-like chest pain, there is no cardiomyopathy or other identifiable organic heart disease or pathological changes in the coronary microcirculation to account for the limited increases in coronary flow in response to vasodilators and pacing stress reported by several groups.61 Thus, many investigators since the 1960s have speculated that disease or dysfunction of the coronary microcirculation may be responsible for angina-like chest pain symptoms and abnormal test results, often referred to as “syndrome X,” in this patient population.63

In recent years, attention has focused on the role of the endothelium in modulating coronary microvascular function, after recognition of the importance of the endothelium in regulating epicardial coronary artery vasomotor tone. Several groups have shown that risk factors for coronary atherosclerosis, including hypercholesterolemia, hypertension, diabetes, and cigarette smoking, can diminish the coronary flow response to endothelium-dependent vasodilators such as acetylcholine in the absence of significant atherosclerosis of epicardial coronary arteries.64–67 Other studies suggest that sympathetic stimuli, such as mental stress, may provoke coronary microvascular constrictor responses.68 In addition, coronary microvascular dysfunction may coexist with atherosclerotic coronary artery disease and contribute to acute and chronic ischemic syndromes,69,70 as has been demonstrated in patients with dilated cardiomyopathy.71

Although these studies support the role of coronary microvascular endothelial dysfunction in explaining chest pain symptoms in patients with normal coronary angiograms or minimal coronary atherosclerosis, convincing evidence of a causal relationship between coronary microvascular dysfunction and myocardial ischemia is lacking at present. In part, such demonstration is hampered by methodological issues regarding measurement of coronary blood flow and detection of mild degrees of myocardial ischemia in humans, as well as by

the absence of normal control subjects who, for ethical reasons, cannot undergo cardiac catheterization. Finally, we strongly emphasize that investigations designed to elucidate the regulation of the coronary microcirculation in health and disease should incorporate the many technological advances of molecular biology, biochemistry, imaging, and modeling with physiological measurements.

Some experimental animal models seem to mimic the clinical picture of syndrome X described above and may be appropriate for investigating mechanisms underlying the microvascular pathology. For example, endothelium-dependent dilation of the coronary microcirculation is impaired in a canine model of diabetes,72 whereas dilation to endothelium-independent agonists is preserved. It has recently been shown that responses of the diabetic coronary microvasculature to hypoperfusion are substantially blunted in dogs with diabetes.73 The mechanisms responsible for these alterations and the influences of other pathological conditions on the coronary microcirculation, eg, hypertension, are not known. In cats with experimental hypertension and left ventricular hypertrophy, coronary microvascular resistance is shifted to the smaller coronary microvessels.74 These results contrast with those seen in the cerebral,75 mesenteric,76 and skeletal77 microvascular beds of rats, in which resistance is shifted upstream. Clearly, species variability needs to be ruled out as a reason for these differences, and definitive studies ultimately must be carried out with human coronary vessels.

The pattern of blood flow to the myocardium is also influenced by coronary collateral vessels. Microvascular arterial anastomoses can enlarge into major conduits that modify the effects of an atherosclerotic obstruction. In human hearts, the distribution and extent of collateral vessels are quite variable. In dogs, native collateral vessels average 40 μm in diameter78 and appear to have little or no function. However, when myocardial perfusion is compromised by obstruction of major vessels, the collaterals enlarge and the blood flow through them increases. Collaterals become angiographically visible only when coronary occlusion is complete or virtually so.79 In the event of a coronary occlusion, collaterals can provide perfusion sufficient to maintain myocardial viability or to prevent myocardial infarction or even sudden ischemic death.80 Many unanswered questions related to the enlargement of existing collaterals and the development of new ones remain. For example, What are the signals that induce coronary collateral development? Are there different signals that initiate the development versus those that continue the growth? What causes growth to stop? All of these questions remain unanswered but are of cardinal importance to the understanding of vascular adaptations to myocardial ischemia.

Regulation of tone in collateral vessels would appear to have importance in modulating blood flow to a collateral-dependent region after a coronary occlusion or severe stenosis. In the beating canine heart, collaterals <100 μm are not maximally dilated immediately after coronary occlusion; rather, this occurs over a period of many minutes.81 This vasodilation appears to involve opening of ATP-sensitive K^+ channels, but many other factors may also play critical roles. Reactivity of collateral vessels to both vasodilators and vasoconstrictors shows similarities to and differences from comparably sized noncollateral vessels.82 These investigations, although crucial to the understanding of vasomotor control of collaterals,

have only begun to examine the contributions of species differences, maturity of collateral vessels, and the inciting stimulus.

Future Directions

Future research needs to encompass both basic and clinical investigations. As indicated throughout the above discourse, there is a great need for inquiry into basic mechanisms at a variety of scientific levels, from integrative techniques of modeling complex systems to reductionist approaches that can decipher complexities of gene expression at all levels of the coronary vasculature. It cannot be emphasized strongly enough that many important fundamental mechanisms, such as metabolic hyperemia, are still poorly understood and that eventual understanding will most likely arise from integrative approaches. Areas such as phenotypic specialization of vascular cells in various sites of the microcirculation and angiogenesis also deserve emphasis, with relevant models. Furthermore, the use of transgenic and gene knockout models has enormous potential for providing fresh insight into the physiology and pathophysiology of the coronary microcirculation. Conversely, clinically oriented investigations will be indispensable to understanding the role of the coronary microcirculation in human disease and to determining whether there is a link between coronary microvascular dysfunction and inducible myocardial ischemia. There is a strong need for further refinement of noninvasive methodologies, such as PET and MRI, that can measure flow and cellular metabolism. These techniques, with higher resolution and greater sensitivity, will be central to documenting clinical consequences of coronary microvascular pathological conditions and establishing the relationship between heterogeneity of flow and metabolism in humans. Therapies that might improve microvascular endothelial vasomotor responsiveness, such as cholesterol reduction, antioxidant therapy, estrogen replacement, L-arginine supplementation, and use of nitric oxide donors, should be tested in patients with evidence of coronary microvascular dysfunction and evidence of myocardial ischemia during stress. The workshop participants perceived that there is an imposing discrepancy between current experimental studies of the coronary microcirculation and the needed clinical investigations in this area and suggest that future initiatives sponsored by the National Institutes of Health should emphasize bridging this gap. Finally, we strongly emphasize that investigations should incorporate the many technological advances of molecular biology in conjunction with physiological measurements to best elucidate the regulation of the coronary microcirculation in health and disease.

Acknowledgments

This workshop was supported by the Division of Heart and Vascular Diseases, National Heart, Lung, and Blood Institute, National Institutes of Health.

Appendix

Appendix Participants in the NHLBI Workshop

William M. Chilian, PhD, Medical Physiology, Texas A&M University, College Station, Tex; James B. Bassingthwaighte, MD, PhD, Center for Bioengineering, University of Washington, Seattle, Wash; Richard O. Cannon III, MD, Cardiology Branch, National

Heart, Lung, and Blood Institute, Bethesda, Md; Fitz-Roy E. Curry, PhD, School of Medicine, University of California, Davis, Calif; Michael J. Davis, PhD, Department of Medical Physiology, Texas A&M University, College Station, Tex; Kevin C. Dellsperger, MD, PhD, Department of Internal Medicine, University of Iowa, Iowa City, Iowa; Brian R. Duling, PhD, Department of Physiology, University of Virginia School of Medicine, Charlottesville, Va; Valentin Fuster, MD, PhD, Cardiovascular Institute, Mount Sinai Medical Center, New York, NY; Mary Ellen Gerritsen, PhD, Miles Institute, West Haven, Conn; Julien I.E. Hoffman, MD, Cardiovascular Research Institute, University of California, San Francisco, Calif; Fumihiko Kajiya, PhD, Department of Medical Engineering and Systems Cardiology, Kawasaki Medical School, Okayama, Japan; David D. Ku, MD, PhD, Department of Pharmacology, University of Alabama at Birmingham, Birmingham, Ala; Kathryn G. Lamping, PhD, Department of Internal Medicine, University of Iowa, Iowa City, Iowa; M. Harold Laughlin, PhD, Veterinary Biomedical Sciences, University of Missouri-Columbia, Columbia, Mo; Erik L. Ritman, MD, PhD, Biodynamics Research Unit, Mayo Medical School, Rochester, Minn; Aubrey E. Taylor, PhD, Department of Physiology, School of Medicine, University of South Alabama, Mobile, Ala; Alfred Small, PhD, Vascular Research Program, National Heart, Lung, and Blood Institute, NIH, Bethesda, Md.

References

1. Nellis SH, Liedtke AJ, Whitesell L. Small coronary vessel pressure and diameter in an intact beating rabbit heart using fixed-position and free-motion techniques. Circ Res. 1981; 49:342–353. [PubMed: 7249271]
2. Yada T, Hiramatsu O, Kimura A, Goto M, Ogasawara Y, Tsujioka K, Yamamori S, Ohno K, Hosaka H, Kajiya F. In vivo observation of subendocardial microvessels of the beating porcine heart using a needle-probe video-microscope with a CCD camera. Circ Res. 1993; 60:100–200.
3. Yada T, Hiramatsu O, Kimura A, Tachibana H, Chiba Y, Lu S, Goto M, Ogasawara Y, Tsujioka K, Kajiya F. Direct in vivo observation of subendocardial arteriolar response during reactive hyperemia. Circ Res. 1995; 77:622–631. [PubMed: 7641332]
4. Hiramatsu O, Goto M, Yada T, Kimura A, Tachibana H, Ogasawara Y, Tsujioka K, Kajiya F. Diameters of subendocardial arterioles and venules during prolonged diastoles in canine left ventricles. Circ Res. 1994; 75:393–397. [PubMed: 8033348]
5. Yada T, Hiramatsu O, Goto M, Ogasawara Y, Kimura A, Yamamoto T, Tsujioka K, Kajiya F. Effects of nitroglycerin on diameter and pulsation-amplitude of subendocardial arterioles in beating porcine heart. Am J Physiol. 1994; 36:H1719–H1725. [PubMed: 7977804]
6. Flannery BP, Dickman HW, Roberge WG, D'Amico KL. Three-dimensional x-ray micro tomography. Science. 1987; 237:1439–1444. [PubMed: 17816787]
7. Avula RTV, Dunsmuir JH, Beighley PE, Thomas PJ, Faridani A, Ritman EL. A micro tomographic technique enhanced by novel image reconstruction algorithm: application to rat coronary vessels. FASEB J. 1994; 8:A854. Abstract.
8. Ritman, EL.; Dunsmuir, JH.; Faridani, A.; Finch, DV.; Smith, KT.; Thomas, PJ. Local reconstruction applied to x-ray tomography. In: Chavent, G.; Papanicolaou, G.; Sacks, P.; Symes, W., editors. IMA Volumes in Mathematics and Its Applications, vol 90: Inverse Problems in Wave Propagation. New York, NY: Springer-Verlag; 1997. p. 443-452.
9. Kuo L, Davis MJ, Chilian WM. Endothelium-dependent, flow-induced dilation of isolated coronary arterioles. Am J Physiol. 1990; 259:H1063–H1070. [PubMed: 2221113]
10. Kuo L, Davis MJ, Chilian WM. Myogenic activity in isolated subepicardial and subendocardial coronary arterioles. Am J Physiol. 1988; 255:H1558–H1562. [PubMed: 2462367]
11. Miller FJ, Dellsperger KC, Embrey RP, Gutterman D. Isolated human coronary arterioles exhibit myogenic tone. Circulation. 1993; 88(suppl I):1–471. Abstract.

12. Yuan Y, Chilian WM, DeFily DV, Granger HJ, Zawieja DC. Permeability to albumin in isolated coronary venules. Am J Physiol. 1993; 265:H543–H552. [PubMed: 8368358]
13. Gerritsen ME, Carley W, Milici AJ. Microvascular endothelial cells: isolation, identification, and activation. Adv Cell Culture. 1988; 6:35–66.
14. Nishida M, Carley WW, Gerritsen ME, Ellingsen O, Kelley RA, Smith TW. Isolation and characterization of human and rat cardiac microvascular endothelial cells. Am J Physiol. 1993; 264:H639–H652. [PubMed: 8447476]
15. Grafe M, Auch-Schwelk W, Graf K, Terbeek D, Hertel H, Unkelbach M, Hildebrandt A, Fleck E. Isolation and characterization of macrovascular and microvascular endothelial cells from human hearts. Am J Physiol. 1994; 267:H2138–H2148. [PubMed: 7810713]
16. Hoffman JIE. Transmural myocardial perfusion. Prog Cardiovasc Dis. 1987; 29:429–464. [PubMed: 2953043]
17. King RB, Bassingthwaighte JB, Hales JRS, Rowell LB. Stability of heterogeneity of myocardial blood flow in normal awake baboons. Circ Res. 1985; 57:285–295. [PubMed: 4017198]
18. Gonzalez F, Bassingthwaighte JB. Heterogeneities in regional volumes of distribution and flows in the rabbit heart. Am J Physiol. 1990; 258:H1012–H1024. [PubMed: 2109937]
19. Bassingthwaighte JB, Malone MA, Moffett TC, King RB, Chan IS, Link JM, Krohn KA. Molecular and particulate depositions for regional myocardial flows in sheep. Circ Res. 1990; 66:1328–1344. [PubMed: 2335030]
20. Austin RE Jr, Aldea GS, Coggins DL, Flynn AE, Hoffman JIE. Profound spatial heterogeneity of coronary reserve: discordance between patterns of resting and maximal myocardial blood flow. Circ Res. 1990; 67:319–331. [PubMed: 2376074]
21. Bassingthwaighte, JB.; Liebovitch, LS.; West, BJ. Fractal Physiology. New York, NY: Oxford University Press; 1994.
22. Bassingthwaighte JB, Van Beek JH, King RB. Fractal branchings: the basis of myocardial flow heterogeneities? Ann N Y Acad Sci. 1995; 591:392–401. [PubMed: 2197931]
23. Coggins DL, Flynn AE, Austin RE Jr, Aldea GS, Muehrcke D, Goto M, Hoffman JIE. Nonuniform loss of regional flow reserve during myocardial ischemia in dogs. Circ Res. 1990; 67:253–264. [PubMed: 2376070]
24. Caldwell JH, Martin GV, Raymond GM, Bassingthwaighte JB. Regional myocardial flow and capillary permeability: surface area products are nearly proportional. Am J Physiol. 1994; 267:H654–H666. [PubMed: 8067421]
25. Hoffman JIE. Heterogeneity of myocardial blood flow. Basic Res Cardiol. 1995; 90:103–111. [PubMed: 7646414]
26. Kuo L, Davis MJ, Chilian WM. Longitudinal gradients for endothelium-dependent and -independent vascular responses in the coronary circulation. Circulation. 1995; 92:518–525. [PubMed: 7543382]
27. Chilian WM. Functional distribution of α_1- and β_2-adrenergic receptors in the coronary microcirculation. Circulation. 1991; 84:2108–2122. [PubMed: 1682067]
28. Jones CJH, DeFily DV, Patterson J, Chilian WM. Endothelium-dependent relaxation competes with α_1- and α_2-adrenergic constriction in the coronary microcirculation. Circulation. 1993; 87:1264–1274. [PubMed: 8384938]
29. Lundberg MS, Sadhu DN, Grumman VE, Chilian WM, Ramos KS. Actin isoform and α_{1b}-adrenoceptor gene expression in aortic and coronary smooth muscle is influenced by cyclical stretch. In Vitro Cell Dev Biol Anim. 1995; 31:115–121. [PubMed: 7735563]
30. Kanatsuka H, Lamping KG, Eastham CL, Dellsperger KC, Marcus ML. Comparison of the effects of increased myocardial oxygen consumption and adenosine on the coronary microvascular resistance. Circ Res. 1989; 65:1296–1305. [PubMed: 2805245]
31. Kuo L, Davis MJ, Chilian WM. Interaction of pressure- and flow-induced responses in porcine resistance vessels. Am J Physiol. 1991; 261:H1706–H1715. [PubMed: 1750529]
32. Kuo L, Arko F, Chilian WM, Davis MJ. Coronary venular responses to flow and pressure. Circ Res. 1993; 72:607–615. [PubMed: 8431988]
33. Kuo L, Davis MJ, Chilian WM. Endothelial modulation of arteriolar tone. News Physiol Sci. 1992; 7:5–9.

34. Xia J, Little TL, Duling BR. Cellular pathways of the conducted electrical response in arterioles of hamster cheek pouch in vitro. Am J Physiol. 1995; 269:H2020–H2038. (Heart Circ Physiol 38).
35. Little TL, Beyer EC, Duling BR. Connexin 43 and connexin 40 gap junctional proteins are present in arteriolar smooth muscle and endothelium in vivo. Am J Physiol. 1995; 268:H729–H739. [PubMed: 7864199]
36. Little TL, Xia J, Duling BR. Dye tracers define differential endothelial and smooth muscle coupling patterns within the arteriolar wall. Circ Res. 1995; 76:498–504. [PubMed: 7859395]
37. Segal S, Beny JL. Intracellular recording and dye transfer in arterioles during blood flow control. Am J Physiol. 1992; 263:H1–H7. [PubMed: 1636748]
38. Little TL, Xia J, Duling BR. Dye tracers define differential endothelial and smooth muscle coupling patterns within the arteriolar wall. Circ Res. 1995; 76:498–504. [PubMed: 7859395]
39. Ku, DD.; Caulfield, JB.; Kirklin, JK. Endothelium-dependent responses in human coronary blood vessels. In: Ryan, U.; Rubanyi, G., editors. Endothelial Regulation of Vascular Tone. New York, NY: Marcel Dekker Inc; 1992. p. 197-223.
40. Ku DD, Liu Q. Species variations in acetylcholine modulation of coronary microvascular reactivity in human, porcine and dog. Can J Physiol Pharmacol. 1994; 72(suppl 4):37. Abstract.
41. Ku DD, Winn MJ, Grigsby T, Caulfield JB. Human coronary vascular smooth muscle and endothelium-dependent responses after storage at −75°C. Cryobiology. 1992; 29:199–209. [PubMed: 1582228]
42. Laughlin MH, McAllister RM. Exercise training-induced coronary vascular adaptations. J Appl Physiol. 1992; 73:2209–2225. [PubMed: 1490929]
43. Laughlin MH, Overholser KA, Bhatte M. Exercise training increases coronary transport reserve in miniature swine. J Appl Physiol. 1989; 67:1140–1149. [PubMed: 2551878]
44. Breisch EA, White FC, Nimmo LE, McKirnon MD, Bloor CM. Exercise-induced cardiac hypertrophy: a correlation of blood flow and microvasculature. J Appl Physiol. 1986; 60:1259–1267. [PubMed: 2939050]
45. Muller JM, Myers PR, Laughlin MH. Exercise training alters myogenic responses in porcine coronary resistance arteries. J Appl Physiol. 1993; 75:2677–2682. [PubMed: 8125889]
46. Muller JM, Myers PR, Laughlin MH. Vasodilator responses of coronary resistance arteries from exercise-trained pigs. Circulation. 1994; 89:2308–2314. [PubMed: 8181157]
47. Sessa WC, Pritchard K, Sevedi N, Wang J, Hintze TH. Chronic exercise in dogs increases coronary vascular nitric oxide synthase gene expression. Circ Res. 1994; 74:349–353. [PubMed: 7507417]
48. Yuan Y, Granger HJ, Zawieja DC, Chilian WM. Flow modulates coronary venular permeability by a nitric oxide-related mechanism. Am J Physiol. 1992; 263:H641–H646. [PubMed: 1510161]
49. Yuan Y, Granger HJ, Zawieja DC, DeFily DV, Chilian WM. Histamine increases venular permeability via a phospholipase C-NO synthase-guanylate cyclase cascade. Am J Physiol. 1993; 264:H1734–H1739. [PubMed: 7684577]
50. Curry FE. Landis Award Lecture: Regulation of water and solute exchange in microvessel endothelium: studies in single capillaries. Microcirculation. 1994; 1:11–26. [PubMed: 8790574]
51. Pagakis SN, Curry FE. Imaging of Ca^{2+} transients in endothelial cells of single perfused capillaries: correlation of peak $[Ca^{2+}]_j$ with site of macromolecular leakage. Microcirculalion. 1994; 1:213–230.
52. He P, Curry FE. Depolarization modulates endothelial cell calcium influx and microvessel permeability. Am J Physiol. 1991; 261:H1246–H1254. [PubMed: 1928406]
53. Curry FE. Modulation of venular microvessel permeability by calcium influx into endothelial cells. FASEB J. 1992; 6:2456–2466. [PubMed: 1563597]
54. He P, Curry FE. Endothelial cell hyperpolarization increases $[Ca^{2+}]_j$ and venular permeability. J Appl Physiol. 1994; 76:2288–2297. [PubMed: 7928849]
55. He P, Curry FE. Differential actions of cAMP on endothelial $[Ca^{2+}]_j$ and permeability in microvessels exposed to ATP. Am J Physiol. 1993; 265:H1019–H1023. [PubMed: 8214108]
56. Kurose I, Anderson DC, Miyasaka M, Tamatano T, Paulson JC, Toda RF, Rusche JR, Granger DN. Molecular determinants of reperfusion-induced leukocyte adhesion and vascular protein leakage. Circ Res. 1994; 74:336–343. [PubMed: 7507416]

57. Moore TM, Khimenko P, Adkins WK, Miyasaka M, Taylor AE. Adhesion molecules contribute to ischemia and reperfusion-induced injury in the isolated rat lung. J Appl Physiol. 1990; 78:2245–2252. [PubMed: 7665425]
58. Ma X-L, Lefer DJ, Lefer AM, Rothlein R. Coronary endothelial and cardiac protective effects of a monoclonal antibody to intercellular adhesion molecule-1 in myocardial ischemia and reperfusion. Circulation. 1992; 86:937–946. [PubMed: 1355412]
59. Ma X-L, Weyrich AS, Lefer DJ, Buerke M, Albertine KH, Kishimoto TK, Lefer AM. Monoclonal antibody to L-selectin attenuates neutrophil accumulation and protects ischemic reperfused cat myocardium. Circulation. 1993; 88:649–658. [PubMed: 7687936]
60. O'Neill P, Charlat M, Michael L, Roberts R, Bolli R. Influence of neutrophil depletion on myocardial function and flow after reversible ischemia. Am J Physiol. 1989; 252:H341–H351. [PubMed: 2916669]
61. Cannon RO, Carnici PG, Epstein SE. Pathophysiological dilemma of syndrome X. Circulation. 1992; 85:883–892. [PubMed: 1537124]
62. Maron BJ, Wolfson JK, Epstein SE, Roberts WC. Intramural ('small vessel') coronary artery disease in hypertrophic cardiomyopathy. J Am Coll Cardiol. 1986; 8:545–557. [PubMed: 3745699]
63. Tanaka M, Fugiwara H, Onodera T, Wu D-J, Matsuda M, Hamashima Y, Kawai C. Quantitative analysis of narrowings of intramyocardial small arteries in normal hearts, hypertensive hearts, and hearts with hypertrophic cardiomyopathy. Circulation. 1987; 75:1130–1139. [PubMed: 3552306]
64. Zeiher AM, Drexler H, Wollschlager H, Just H. Modulation of coronary vasomotor tone in humans: progressive endothelial dysfunction with different early stages of coronary atherosclerosis. Circulation. 1991; 83:391–401. [PubMed: 1991363]
65. Egashira K, Inov T, Hirooka Y, Yamada A, Maruoka Y, Kai H, Svgimachi M, Suzuki S, Takeshita A. Impaired coronary blood flow response to acetylcholine in patients with coronary risk factors and proximal atherosclerotic lesions. J Clin Invest. 1993; 91:29–37. [PubMed: 8423226]
66. Quyyumi AA, Dakak N, Andrews NP, Husain S, Arora S, Gilligan DM, Panza JA, Cannon RO. Nitric oxide activity in the human coronary circulation: impact of risk factors for coronary atherosclerosis. J Clin Invest. 1995; 95:1747–1755. [PubMed: 7706483]
67. Quillen JE, Rossen JD, Oskarsson HJ, Minor RL, Lopez AG, Winnford MD. Acute effect of cigarette smoking on the coronary circulation: constriction of epicardial and resistance vessels. J Am Coll Cardiol. 1993; 22:642–647. [PubMed: 8354792]
68. Dakak N, Quyyumi AA, Eisenofer G, Goldstein DS, Cannon RO. Sympathetically mediated effects of mental stress on the cardiac microcirculation of patients with coronary artery disease. Am J Cardiol. 1995; 76:125–130. [PubMed: 7611145]
69. Wilson RF, Johnson MR, Marcus ML, Aylward PE, Skorton DJ, Collins S, White CW. The effect of coronary angioplasty on coronary flow reserve. Circulation. 1998; 77:873–885. [PubMed: 2964950]
70. Oren NG, Crake T, Lefroy DC, De Silva R, Davies GJ, Maseri A. Reduced coronary vasodilator function in infarcted and normal myocardium after myocardial infarction. N Engl J Med. 1994; 331:222–227. [PubMed: 7832835]
71. Cannon RO, Cunnion RE, Parrillo JE, Palmeri ST, Tucker EE, Schenke WH, Epstein SE. Dynamic limitation of coronary vasodilator reserve in patients with dilated cardiomyopathy and chest pain. J Am Coll Cardiol. 1987; 10:1190–1200. [PubMed: 3680786]
72. Dellsperger KC, Brooks LA, Eastham CL, Guttermann DD. Coronary microvascular vasodilator responses in diabetic dogs. Circulation. 1991; 84(suppl II) II-558. Abstract.
73. Kersten JR, Brooks LA, Dellsperger KC. Impaired microvascular responses to graded coronary occlusion in diabetic and hyperglycemic dogs. Am J Physiol. 1995; 268:H1667–H1674. [PubMed: 7733369]
74. Kanatsuka H, Lamping KG, Eastham CL, Marcus ML, Dellsperger KC. Coronary microvascular resistance in hypertensive cats. Circ Res. 1991; 68:726–733. [PubMed: 1742865]
75. Baumbach GL, Dobrin PB, Hart MN, Heistad DD. Mechanics of cerebral arterioles in hypertensive rats. Circ Res. 1988; 62:56–64. [PubMed: 3335057]

76. Bohlen HG. Intestinal microvascular adaptation during maturation of spontaneously hypertensive rats. Hypertension. 1983; 5:739–745. [PubMed: 6618636]

77. Wiegman DL, Joshua IG, Morff RJ, Harris PD, Miller FN. Microvascular responses to norepinephrine in renovascular and spontaneously hypertensive rats. Am J Physiol. 1979; 236:H545–H548. [PubMed: 434218]

78. Schaper, W. The morphology of collaterals and anastomoses in human, canine and porcine hearts. In: Schaper, W.; Schaper, J., editors. The Collateral Circulation of the Heart. New York, NY: Elsevier; 1971.

79. Schaper, W. Comparative arteriography of the collateral circulation. In: Schaper, W.; Schaper, J., editors. The Collateral Circulation of the Heart. New York, NY: Elsevier; 1971.

80. Marcus, ML. The coronary collateral circulation. In: Schaper, W.; Schaper, J., editors. The Coronary Circulation in Health and Disease. New York, NY: McGraw-Hill; 1983.

81. Lamping KG, Bloom EN, Harrison DG. Regulation of native collateral vessel dilation after coronary occlusion in the dog. Am J Physiol. 1994; 266:H769–H778. [PubMed: 8141378]

82. Lamping KG, Eastham CL, Bloom EN. Responses of coronary microvascular collaterals to endothelin and vasopressin in vivo. Circulation. 1993; 88(suppl I) I-375. Abstract.