Integrated regulation of pressure and flow in the coronary microcirculation

Judy M. Muller, Michael J. Davis, William M. Chilian *

Microcirculation Research Institute, Department of Medical Physiology, Texas A and M, University Health Science Center, College Station, TX, USA
Department of Physiology, Medical College of Wisconsin, 8701 Watertown Plank Road, Milwaukee, WI 53226-0509, USA

Received 12 December 1995: accepted 13 March 1996

Abstract

There are many mechanisms that contribute to the regulation of coronary blood flow including vasodilatory metabolites, myogenic regulation, flow- or shear stress-mediated vasodilation, and neurohumoral influences. It is interesting to note that coronary arterioles of varying sizes appear to possess different sensitivities to these regulatory factors, but each appears to dominate the control of a particular segment of the microvasculature. For example, during metabolic hyperemia the smallest arterioles seem to be most sensitive to the effects of metabolites, but metabolic and myogenic mechanisms that dictate the tone of upstream microvessels likely act in concert to facilitate the overall adjustments in coronary vasomotor tone. Neurohumoral mechanisms seem to modulate the robustness of these intrinsic adjustments, perhaps by restraining the extent of vasodilation. The purpose of this review is to discuss these many regulatory mechanisms and also present a framework by which the vasoactive reactions elicited by these different mechanisms are integrated into coordinated responses of the entire coronary vascular network.

Keywords: Coronary vasculature; Microcirculation; Adenosine; Endothelium; Vascular conductance; Autonomic nervous system

1. Introduction

The mechanisms which contribute to control of pressure and blood flow in the coronary circulation must serve two purposes. First, blood flow must be suited to the metabolic demands of the myocardium. That is, blood flow must be able to change in parallel with changes in metabolic demand. Secondly, capillary pressure must be regulated and maintained relatively constant despite changes in perfusion pressure so that efficient delivery of oxygen and nutrients is ensured without producing damage to exchange vessels. The majority of coronary vascular resistance which serves to control both local blood flow and capillary perfusion pressure is found in coronary arterioles less than 150 μm in diameter [1]. However, even within this section of the coronary vasculature which is often considered collectively as the ‘resistance vasculature’, heterogeneity of function exists. Considerable data now exist which suggest that vascular reactivity differs between large and small coronary arterioles and that vasoactive reactions from similarly sized endocardial and epicardial arterioles also differ. Recent information concerning the role of venules in control of coronary microvascular resistance has also come to light. Thus regional and positional differences must be considered when pressure/flow relationships and the control of resistance within the coronary microcirculation are investigated. These differences are often highlighted by the heterogeneous effects exerted on the coronary microcirculation during pathological conditions. An example of this selectivity is the susceptibility of the left subendocardium to ischemia [2]. This particular observation emphasizes that one must exercise caution when extrapolating observations from the behavior of subepicardial microvessels to those in the subendocardium. We have focused our discussion on the subepicardial microcirculation merely because the bulk of work has been performed in vessels from this area. Taken together, the preceding discussion reinforces the idea that the microcirculation is not a homogenous regulatory unit, rather there are many
differences in regulation of tone throughout the coronary vasculature. This emphasizes the need to view the coronary microcirculation as an integrated network of microvascular domains which may show independent, yet coordinated, responses to global stimuli.

The purpose of this review is to summarize the current knowledge of the mechanisms which contribute to regulation of microvascular resistance in the coronary circulation. The segmental distribution of the regulatory mechanisms will be emphasized and we will speculate about how these discrete microvascular regions interact to provide efficient distribution of blood flow to the entire myocardium.

2. Distribution of coronary microvascular resistance

The distribution of resistance in the coronary circulation is reflected by the profile of microvascular pressures. Resistance of any segment is directly proportional to the magnitude of the pressure drop across that segment. Information about the distribution of resistance also provides a perspective for the analysis of regulatory mechanisms in different sizes of microvessels because it engenders insight into the impact a change in microvascular caliber has on total coronary resistance. Specifically, if a high-resistance site (e.g., arterioles) constricts by 20%, the repercussions on coronary resistance will be greater than if a similar change in diameter occurred in a low-resistance site. In the microcirculation of the heart there are a few reports that define the locations of resistance in the epicardium and endocardium.

Nellis and colleagues [3] found that in the beating right ventricle of the rabbit approximately 30% of the total pressure dissipation occurred in arterial vessels proximal to 140 μm in diameter. Chilian and colleagues made similar observations in the feline left ventricle with about 20% of the pressure loss occurring proximal to 200 μm small arteries and another 20% between 100 and 200 μm arterial microvessels [1]. Both of these studies support the idea that a significant proportion of coronary resistance can be attributed to arterial microvessels greater than 100 μm in diameter. Tillmanns et al. [4], however, reported there was almost no pressure drop from the aorta to vessels of this size in the rat heart. The reasons for the differences in the proportion of resistance residing in relatively large microvessels among the above referenced reports are not...
obvious, but are likely related to the preparation or species. Despite the differences in the contribution of small arteries to coronary vascular resistance, we emphasize that all of the reports found that arterioles constitute the major fraction of coronary resistance.

The site of active coronary resistance (that modulated by the vasodilators, dipyridamole and papaverine) has been examined by Chilian et al. [1,5]. Fig. 1 portrays the distribution of microvascular pressures under conditions with intact coronary vasomotor tone and during intense vasodilation with dipyridamole (sufficient to increase total flow 5- to 6-fold). Dipyridamole reduced pressures in small arteries vessels less than 170 μm in diameter while increasing venular pressures [5]. During the hyperemia, the pressure drop across the microcirculation decreased by more than 50% (from 50 to 20 mmHg). Resistance of this microvascular segment, based on the quotient of the pressure drop and flow, was decreased by about 12- to 16-fold. Resistance of the arterial and venous compartments during administration of dipyridamole also revealed markedly heterogeneous changes; resistance of small arteries decreased by 33% and venous resistance was unchanged. Less intense vasodilation by papaverine appeared to preferentially vasodilate small arteries, because arteriolar pressures increased after administration of this compound. In the aggregate, these findings point to two important attributes related to the control of coronary resistance. First, the significant proportion of resistance that resides in small arteries is, in part, due to active mechanisms, and may, therefore, be modulated by vasodilators and constrictors. Secondly, arterioles have the greatest magnitude of active tone, and this segment may be substantially affected by intense vasodilation.

Coronary microvascular resistance also appears to vary across the wall of the left ventricle [6]. Fig. 2 shows microvascular pressures obtained from similarly-sized microvessels in the subendocardium and subepicardium of the porcine left ventricle. Microvascular pressures were measured during variations in coronary perfusion pressure from 40 to 100 mmHg during maximal vasodilation in an arrested heart that had been incised to expose the endocardial surface. At each level of perfusion pressure, arteriolar pressures in the endocardium were lower than those in the epicardium. Also, endocardial venular pressures were higher than those in epicardial venules. These results suggest that transmural arterioles which course through the ventricular wall and are upstream from the endocardial microcirculation contribute significantly to the total endocardial resistance, as indicated by the lower pressures in the endocardial vs. epicardial arterioles. The results also suggest that microvascular resistance in the subendocardium is significantly lower than that in the subepicardium, because the arteriolar-venular pressure difference is much smaller. Although these results were obtained in a preparation that cannot be described as purely physiological, they do suggest that the distribution of coronary microvascular resistance varies across the wall of the heart.

One additional factor that may play an important role in the determination of transmural variations in resistance are the compressive forces imposed on microvessels in different layers of the left ventricle. Yada and colleagues [7] have made observations of the excursions in diameter that occur during the cardiac cycle in subepicardial and subendocardial microvessels. Compressive forces were much more evident in subendocardial vessels because their diameters were reduced during systole between 10 and 20%, whereas those in the subepicardium were reduced between 0 and 5%. Because resistance is proportional to diameter or radius to the fourth power, these differences must be translated into transmural variations in resistance that occur during a single cardiac cycle.

3. Autoregulation

Coronary autoregulation refers to the intrinsic ability of the heart to maintain blood flow relatively constant despite marked changes in perfusion pressure [8,9]. During conditions of reduced perfusion pressure, vasodilation produced by autoregulatory adjustments contributes to maintenance of adequate blood flow to the myocardium. Autoregulatory
control may also help to prevent edema and microvascular damage by maintaining capillary hydrostatic pressure relatively constant despite large increases in coronary perfusion pressure. Reductions in perfusion pressure produce significant dilation of coronary arterioles smaller than 100 to 150 μm in diameter [10, 11], the magnitude of which is inversely proportional to vessel diameter. These observations indicate that autoregulatory mechanisms, as might be expected from pressure profiles of the coronary circulation, predominate in the microcirculation where they have the greatest impact on total coronary vascular resistance.

3.1. Myogenic responses

In the coronary circulation, the most compelling evidence that a myogenic component contributes to autoregulation has come from studies of isolated coronary arterioles. Because isolated vessels can be removed from the surrounding myocardium, the many variables which may affect resistance to blood flow (thus modulating the myogenic response) can be eliminated or controlled. Kuo et al. [12] demonstrated that porcine coronary arterioles 80 to 100 μm in diameter exhibit myogenic responses to increases and decreases in transmural pressure. Fig. 3 illustrates the evidence that active myogenic responses were present in both subendocardial and subepicardial arterioles. Interestingly, subepicardial arterioles, when compared to subendocardial arterioles, exhibited greater vasodilatory responses at low pressures and augmented constriction at higher pressures. These differences in myogenicity may contribute to transmural differences in autoregulation; specifically, autoregulation in the subepicardium occurs at both lower and higher pressures than in the subendocardium [13]. Kuo et al. [14] also found active myogenic responses of isolated coronary venules. However, these responses differed from those of arterioles, being far less robust. In arterioles, the magnitude of the myogenic constriction produced by increases in intraluminal pressure was sufficient to reduce arteriolar diameter to a value lower than the value determined before the increase in pressure. In venules, a myogenic constriction countered the passive increase in diameter that occurred as luminal pressure was elevated, but did not result in a net decrease in venular diameter following an increase in pressure. These differences in myogenicity may be indicative of the differences in vascular reactivity which exist throughout the coronary microcirculation. From these reports one might conclude that coronary arterioles less than 100 μm in diameter are of primary importance in autoregulation of the coronary microcirculation due to their myogenic capacity. Whereas myogenic responses of coronary arterioles may contribute importantly to autoregulation, alternative autoregulatory mechanisms may exist in other segments of the microvasculature.

3.2. Metabolic responses

Although coronary arterioles display active myogenic responses, metabolic control likely also contributes to, and probably dominates, coronary autoregulation. Even in the absence of alterations in metabolic demand it is possible that changes in perfusion pressure alter the concentration of vasoactive metabolites. For example, a decrease in perfusion pressure would initially cause a reduction in inflow, which could decrease the washout of metabolites. This would lead to a buildup of vasoactive metabolites, thus producing autoregulatory vasodilation. In the intact coronary vasculature it is difficult to separate the mechanisms which contribute to autoregulatory adjustments of blood flow. There is evidence for metabolic induction of autoregulatory responses, however, specific metabolites...
and cellular mechanisms which give rise to metabolic vasodilation during conditions of reduced perfusion pressure remain to be defined.

Some evidence now exists which supports a role for ATP-sensitive potassium channels in mediating changes in coronary blood flow that occur in response to pressure changes within the autoregulatory range and in response to occlusion. Narishige et al. [20] reported that glibenclamide inhibited coronary autoregulatory responses in anesthetized dogs. Komaru et al. [16] found that dilation of canine epicardial arterioles which occurred in response to reductions in perfusion pressure was inhibited by treatment with glibenclamide, an inhibitor of ATP-sensitive potassium channels. Kanatsuka et al. [17] showed that glibenclamide inhibited vasodilatory responses of arterial microvessels (large and small arterioles) during coronary occlusion and during the reactive hyperemia which occurred upon release of the occlusion. In contrast, Kersten et al. [18] reported that in diabetic dogs in which vasodilation of coronary microvessels to reductions in perfusion pressure was significantly impaired, dilation to aprakalin, an ATP-sensitive channel agonist, was slightly enhanced. The results of Kersten and colleagues indicate that an alternative mechanism which contributed to dilation in response to decreases in perfusion pressure was impaired by diabetes whereas the function of the ATP-sensitive potassium channels was actually enhanced, thus suggesting involvement of a mechanism other than ATP-sensitive potassium channels. While it is likely that these potassium channels play a role in autoregulation of coronary blood flow, results of these studies must be interpreted cautiously in light of recent reports showing that glibenclamide has a partial blocking effect on calcium-activated potassium channels and L-type calcium channels [19].

4. Adrenergic control

4.1. α-Adrenergic responses

Although adrenergic innervation is present throughout the coronary microcirculation, a heterogeneity of adrenergic function exists between segments of the coronary microvasculature [20,21]. Large epicardial coronary arteries constrict in response to α,-receptor agonists [22]; however, the primary sites of α-adrenergic regulation in the coronary circulation appear to be in small arteries and large arterioles. In contrast to large epicardial arteries, there appears to be a functional distribution of both α₁-adrenergic and α₂-adrenergic receptors in coronary resistance vessels [20,22]. Activation of α₁-adrenergic receptors by exogenous norepinephrine infusion produces constriction of coronary arterioles greater than 100 μm in diameter and a simultaneous dilation of arterioles less than 100 μm in diameter [20,21]. These diverse effects are likely related to pressure changes created by the constriction of larger arterioles. If the upstream vessels (>100 μm) constrict, this will reduce pressure in the smaller downstream arterioles. The decrease in pressure may then trigger a myogenic dilation in these smaller vessels. Under physiological conditions (i.e., perfusion pressure within the physiological range and intact autoregulatory mechanisms), infusion of the α₁-adrenergic agonist, phenylephrine, produced constriction of small coronary arteries (>100 μm) but did not affect diameters of coronary arterioles (<100 μm). The selective α₁-adrenergic agonist, BHT-933, did not alter diameters of either large or small coronary microvessels. In contrast, if perfusion pressure was maintained below physiological levels (hypoperfusion), autoregulatory responses to pressure changes were eliminated and both α₁- and α₂-adrenergic vasoconstriction were unmasked throughout the coronary microcirculation [20].

α-Adrenergic vasoconstriction is modulated by endogenous mechanisms. First, as mentioned in the preceding discussion, microvascular constriction is augmented during hypoperfusion. Second, blockade of adenosine receptors also appears to differentially modulate α₁-adrenergic and α₂-adrenergic constriction. DeFily et al. [23] demonstrated that endogenous adenosine modulates α₂-adrenergic constriction of epicardial arterioles. Under baseline conditions, α₂-adrenergic activation did not alter diameters of epicardial arterioles; however, administration of the adenosine antagonist, 8-p-sulfophenyltheophylline, unmasked significant constriction of arterioles upon α₂-adrenergic stimulation. α₁-Adrenergic constriction was unaffected by adenosine antagonism. Jones et al. [24] found that vasoconstrictor responses of small epicardial arteries and arterioles to α₁- and α₂-agonists were potentiated by the L-arginine analogs, N⁷-nitro-L-arginine and N⁷-nitro-L-arginine-methyl ester, suggesting that endothelium-dependent release of nitric oxide occurs simultaneously with α-adrenergic activation and may compete with the adrenergic constriction. Under most conditions, intrinsic vasodilatory escape from α-adrenergic constriction appears to compensate for undue constrictor effects, however, in pathological conditions that impair endothelium-dependent vasodilatory responses, α-adrenergic constrictor responses may be exaggerated.

Functional responses to α-adrenergic stimulation have also been examined in isolated coronary microvessels. Interestingly, although the effects of α-adrenergic stimulation on coronary arteries and arterioles have been well documented in vivo, isolated coronary arterioles do not respond to α-adrenergic agonists. Isolated coronary resistance arteries do not constrict in response to either epinephrine or norepinephrine, even in the presence of propranolol [25]. α-Adrenergic activation by norepinephrine neither constricted isolated arterioles (approximately 100 μm in diameter) nor attenuated the dilation of arterioles to adenosine [26]. In contrast, isolated coronary venules of similar size (100 μm diameter) constricted in response to norepinephrine [26]. This vasoconstrictor re-
response could be blocked by either prazosin, an α₁-adrenergic antagonist, or by rauwolscine, an α₂-adrenergic antagonist, indicating the presence of both receptor subtypes in coronary venules. The reasons which underlie the failure of isolated coronary arterioles to constrict in response to adrenergic agonists remain to be ascertained. It is possible that the lack of responsiveness found in the isolated vessels is related to the isolation procedure. However, isolated venules and skeletal muscle artery arterioles which were handled in a similar manner displayed constriction to norepinephrine. It is also possible that the constrictor responses documented in in-vivo preparations actually occurred as a secondary effect of adrenergic stimulation rather than as a direct coupling of adrenergic stimulation and receptor activation. For example, adrenergic constriction of venules could cause an increase in upstream pressure initiating a myogenic constriction of arterioles. Adrenergic stimulation could also lead to production of a vasoconstrictor in parenchymal tissue in vivo; such an effect would be lost in an isolated arteriole. These possibilities remain to be investigated and questions such as this confirm the need for techniques in which the many control factors present in the coronary microcirculation can be manipulated independently.

4.2. β-Adrenergic responses

Whereas a great deal of information exists regarding the role of α-adrenergic responses in the coronary microcirculation, far less is known concerning microvascular responses to β-adrenergic activation. β-Adrenergic receptors have been identified in the coronary vasculature [27]. Activation of both β₁-adrenergic and β₂-adrenergic receptors has been shown to elicit vasodilation in the coronary circulation. In general, the vasodilation that occurs due to direct stimulation of coronary vascular β-adrenergic receptors has not been readily distinguishable from the indirect metabolic vasodilation that occurs when heart rate and myocardial contractility are stimulated by cardiac myocyte β₁-adrenergic receptors. Most work that demonstrates a role for vascular β-adrenergic receptors in determining coronary vascular resistance has shown an effect in large arteries or has indirectly shown that resistance vessels are sensitive to β-adrenergic activation. This indirect evidence comes from studies in which changes in coronary vascular resistance have been calculated from pressure/flow measurements in the whole heart under conditions in which myocardial metabolism has been controlled. To eliminate metabolic effects of β-adrenergic activation, Trivella et al. [28] studied isoproterenol-induced vasodilation during prolonged asystoles. Dilation to isoproterenol was blocked by the β₂-selective antagonist, L18,551 and by the β₁-selective agonists, proctolol and L650,744. Radioligand binding studies indicate that as with α-adrenergic receptors, the distribution of β adrenergic receptors is heterogeneous within the coronary circulation. The ratio of β₁:β₂ adrenergic receptors in large vessels is approximately 1.5–2:1 [27] whereas a predominance of β₂-adrenergic receptors is found in resistance vessels [29]. These studies indicate that both β₁-adrenergic and β₂-adrenergic receptors are present in the coronary circulation and can be activated to produce significant vasodilation. However, further work which targets microvascular responses is necessary to determine whether β-adrenergic effects are heterogeneous within the coronary microcirculation. Isolated resistance arteries relax in response to both norepinephrine and isoproterenol [25]; however, the effects of β-adrenergic agonists on microvessels from distinct segments of the coronary microvasculature have not been examined. Therefore, it is not yet known whether β-adrenergic effects differ between arterioles and small arteries or between epicardial and endocardial vessels.

5. Metabolic control

Local metabolic control is thought to be the most important mechanism by which increases in myocardial metabolic demand and oxygen consumption are matched by increases in coronary blood flow, however, the precise metabolic mediators remain to be defined. Although an increase in metabolic activity undoubtedly produces an increase in vasoactive metabolites at all levels of the coronary vasculature, metabolic control mechanisms appear to be most prominent in small coronary arterioles. When the effect of short coronary occlusions was studied in canine epicardial microvessels, only vessels less than 100 μm in diameter dilated during the period of occlusion, whereas both large and small microvessels dilated during the reactive hyperemia [17]. The adenosine antagonist, 8-phenyltheophylline, attenuated the dilation of arterioles less than 100 μm in diameter during the occlusion and inhibited the dilation of both large and small vessels during the reactive hyperemic response. Kanatsuka et al. used rapid pacing and endogenous or exogenous adenosine to produce comparable increases in coronary flow [30]. These investigators found that the changes in coronary microvascular resistance differed with rapid pacing and augmentation of adenosine. Rapid pacing produced vasodilation of both large and small coronary arterioles, whereas both endogenous and exogenous adenosine caused dilation only in small arterioles. These studies suggest that small coronary microvessels are the primary target for the effects of the metabolic vasodilator, adenosine. However, the fact that all segments of the coronary microcirculation dilated during rapid pacing and reactive hyperemia implies that other mechanisms are involved in the vasodilatory responses which accompany changes in metabolic supply and demand. Jones et al. also found that pacing-induced increases in myocardial oxygen demands produced dilation throughout the coronary microvasculature [31]. Of note.
both endogenous and exogenous adenosine produce significant vasodilation of small coronary arterioles in vivo.

Isolated vessel studies also indicate that the vasodilatory effects of adenosine are greatest in small arterioles. Kuo et al. studied vasodilatory responses to adenosine in 4 sizes of arterial vessels [33]. Their study showed that small arterioles were most sensitive to adenosine. Furthermore, adenosine was a more potent dilator of small (25 to 45 μm) and intermediate arterioles (50 to 80 μm) than of large arterioles (90 to 130 μm) and small arteries (140 to 300 μm). Quillen and Harrison [34] studied adenosine relaxation in epicardial and endocardial microvessels ranging in size from 80 to 200 μm in diameter and found endocardial vessels to be more sensitive to adenosine. Piana et al. [35] also investigated vasodilation to adenosine in epicardial and endocardial vessels of similar size (130 to 210 μm) but failed to find any differences in the vasodilatory responses between the two groups. The reasons for these disparate results are not clear. Both studies were performed in porcine vessels, but there were some differences in techniques used. In the study of Quillen and Harrison [34] mature pigs were used and the vessels were preconstricted with the thromboxane analog, U46619, while Piana et al. [35] used young pigs (9–12 weeks) and preconstricted the vessels with acetylcholine. It is worth noting that in both studies the vasodilatory responses to other agonists (bradykinin, A23187, and sodium nitroprusside) were similar in endocardial and epicardial vessels and the sensitivity to these agonists was similar in both studies. Collectively, these results indicate that the sensitivity to adenosine varies between segments of the coronary microcirculation and, therefore, suggest that the importance of adenosine as an endogenous vasodilator may vary depending upon the precise region being studied. Another interesting aspect of the data from Kuo and colleagues [33] pertains to the sensitivity of isolated arterioles to adenosine compared to estimates of the interstitial concentrations of the purine. Many laboratories have estimated the interstitial concentration of adenosine in the range of 4 μM to 50–100 nM [36–38]. The data of Kuo and colleagues [33] would predict that the arterioles would be virtually maximally dilated with such high interstitial concentrations, but in the intact heart there is considerable vasodilatory reserve in these small resistance vessels. Although these in vitro results of arteriolar sensitivity to adenosine appear to be in conflict with the estimates of interstitial levels, we can only speculate that there are other endogenous factors that lessen the sensitivity of arterioles in vivo to the vasodilatory effects of adenosine. Another way to reconcile the results is that the estimates of interstitial concentrations of adenosine may be too high. Despite these apparent conflicts between in vitro sensitivity and estimates of interstitial concentrations, we best summarize by stating that adenosine is a very potent vasodilator in the coronary circulation, and has the potential to contribute to the regulation of coronary blood flow.

Activation of ATP-sensitive potassium channels may also contribute to metabolic regulation of microvascular resistance which occurs in response to changes in metabolic supply/demand. Daut et al. showed that glibenclamide, a pharmacological blocker of ATP-sensitive channels, attenuated hypoxia-induced vasodilation of the coronary circulation [39]. Glibenclamide also prevented coronary blood flow from increasing in response to an increase in myocardial metabolism produced by β1-adrenergic stimulation [40]. However, these studies did not target the microcirculation. The role of these ATP-sensitive potassium channels in mediating vasodilation of the coronary microcirculation during conditions of increased metabolic demand remains to be investigated. Kuo et al. recently reported that adenosine augmented flow-induced vasodilation of isolated coronary arterioles through activation of ATP-sensitive potassium channels [41]. Both adenosine-induced and flow-induced vasodilation have been shown to participate in functional hyperemia [42,43]. This report indicates that these two control mechanisms interact in an additive manner to produce dilation, suggesting that in the intact coronary microcirculation these mechanisms could combine to increase coronary blood flow under conditions of increased myocardial demand.

6. Endothelial modulation of coronary microvascular resistance

The importance of the endothelium in modulating coronary microvascular resistance to blood flow has become increasingly apparent. The endothelium modulates smooth muscle tone through the release of both vasodilators and vasoconstrictors. Endothelium-dependent release of factors such as nitric oxide, prostaglandins, endothelin, and endothelium-derived hyperpolarizing factor may occur in response to humoral substances, changes in intravascular flow (i.e., shear stress), and possibly neural stimulation. Within the coronary circulation, there is now a plethora of evidence to suggest that coronary vascular tone is constantly being modulated by the endothelium under varying physiological conditions. In addition to mediating vasodilatory responses to neurohumoral agents such as serotonin [44,45] and acetylcholine [46], endothelium-dependent modulation of autoregulation [47], α-adrenergic constriction [24], and both reactive hyperemic [48–52] and functional hyperemic [53,54] responses have been demonstrated in the coronary circulation.

A recent study of the microcirculation showed that inhibition of nitric oxide synthesis by Nω-nitro-L-arginine methyl ester (L-NAME) caused distinctly heterogeneous changes in diameter in coronary microvessels. Microvascu-
lar responses to adenosine and pacing were studied before and after administration of L-NAME [31]. In this study, adenosine dilated both small arteries and arterioles and increased coronary blood velocity in the absence of nitric oxide synthesis inhibition. Similarly, before inhibition of nitric oxide synthesis, left atrial pacing caused dilation of both large and small microvessels and increased coronary blood velocity. Inhibition of nitric oxide synthesis produced constriction of small arteries and dilation of arterioles. The dilation of arterioles was likely an autoregulatory response initiated by the constriction of upstream vessels rather than a direct vasodilatory effect of the nitric oxide synthase inhibitor. After inhibition of nitric oxide synthesis, adenosine failed to produce further dilation of the arterioles which had already dilated upon treatment with L-NAME. Thus, following L-NAME treatment the adenosine-induced increase in coronary blood velocity was attenuated. Pacing also failed to produce dilation of coronary microvessels following inhibition of nitric oxide synthesis and the increase in coronary blood velocity was attenuated. These results suggest that endothelium-dependent nitric oxide production contributes to increases in blood flow which occur in response to increased metabolic demand. The heterogeneous changes in baseline microvascular diameter produced by L-NAME may in part explain the lack of effect of nitric oxide synthesis inhibition found in studies in which global coronary blood flow was measured [47,51,54]. Inhibition of nitric oxide synthesis may alter the distribution of coronary resistance, which can alter responses to pharmacological and physiological stimuli (please refer to Lefer and Lefer in this Issue who discuss this topic in the context of reperfusion injury).

Neurohumoral responses in the coronary microcirculation are also subject to modulation by the endothelium. In the intact coronary microcirculation, Jones et al. [24] found that vasoconstrictor responses to α₁- and α₂-adrenergic agonists were attenuated by endothelium-dependent release of nitric oxide. In isolated porcine coronary resistance arteries [55] and arterioles [56], antagonists of nitric oxide synthase produce constriction under baseline conditions, suggesting that there is tonic release of nitric oxide in small coronary vessels. Clonidine [57], serotonin [57], and ADP [44] produce endothelium-dependent relaxation of isolated porcine coronary microvessels through release of nitric oxide. In dogs, dilation of coronary resistance arteries to acetylcholine is also mediated through release of nitric oxide [46]. In the pig, where acetylcholine produces constriction of coronary microvessels, this response is modulated by release of nitric oxide from the endothelium [55]. These data further emphasize the modulatory effect exerted by endothelium-derived nitric oxide on various control mechanisms in the coronary microcirculation.

6.1. Flow-induced vasodilation

Kuo et al. [58] have demonstrated that coronary arterioles exhibit flow-induced vasodilation. Using an in vitro preparation, responses to graded increases in intraluminal flow were evaluated in the absence of pressure changes. This was accomplished by cannulating isolated coronary microvessels with micropipettes that were attached to separate pressure reservoirs. Adjustment of the heights of these reservoirs in equal and opposite directions allowed for establishment of a pressure gradient across the vessel, thus generating flow through the vessel without changing mean intraluminal pressure. Incremental increases in flow produced graded dilation; diameter increased as much as 30% at the highest flow rates. Kuo et al. [58] further showed that flow-induced vasodilation of coronary arterioles is abolished by removal of the endothelium. Flow-induced vasodilation was abolished by inhibition of nitric oxide synthesis with an L-arginine analog, L-NMMA, the effects of which were reversed by addition of excess L-arginine [58]. These experiments indicated that flow-induced vasodilation of coronary arterioles occurred through endothelium-dependent release of nitric oxide. Although the role of flow-mediated vasodilation has not been demonstrated in vivo, Kuo et al. demonstrated that vasodilatory responses to flow and myogenic responses to pressure interact locally in isolated arterioles in both an additive and competitive manner [56]. In arterioles which demonstrated myogenic dilation, diameter increased further upon exposure to intraluminal flow. Alternatively, flow-induced vasodilation of isolated coronary arterioles opposed myogenic constriction to an increase in intraluminal pressure. These findings illustrate the potential importance of flow-induced vasodilation as a regulatory mechanism which could augment, for example, metabolic vasodilation, or conversely, oppose autoregulatory responses.

More recent studies have shown that vasodilatory responses to flow are heterogeneous within the coronary microcirculation. Kuo et al. studied flow-induced vasodilation in 4 sizes of coronary arterial microvessels [33]. Fig. 4 shows that the magnitude of flow-induced vasodilation was greatest in large arterioles (90 to 130 μm). The response to flow was reduced in intermediate arterioles (50 to 80 μm) compared to large arterioles. Small arterioles (25 to 40 μm) were even less responsive to flow than were intermediate arterioles. Small arteries displayed responses to flow similar to those present in the small arterioles. Flow-induced vasodilation also occurred in isolated coronary venules [14]. The vasodilatory responses of venules to flow were, however, not as vigorous as those documented in coronary arterioles. As with arterioles, flow-induced venular dilation was abolished by removal of the endothelium. Interestingly, in venules, removal of the endothelium not only abolished flow-induced vasodilation but resulted in constriction upon exposure to flow. This suggests that removal of the endothelium unmasked a flow-induced constrictor mechanism which was not present in coronary arterioles. Collectively these studies indicate that, similar to other regulatory mechanisms, the influence of flow varies between segments of the coronary microcirculation.
A segmental gradient for flow-induced responses may facilitate coordination of blood flow between segments of the coronary microcirculation.

7. Integration of microvascular control mechanisms

Many different regulatory factors influence the caliber of coronary arterioles and small arteries. Each of these factors appears to have the potential to affect total coronary vascular resistance. Despite knowledge about these various factors, it becomes essential to understand how they interact and compensate for one another. An important facet of interactions among the various regulatory factors relates to the manner by which they act in a concerted integrative manner to maintain proper oxygen delivery to the myocardium. Fig. 5 illustrates a simplified scheme in which metabolic, myogenic, and flow-dependent mechanisms are integrated to optimize the network responses to alterations in metabolism [39]. This scheme is speculative and is likely too simple, inasmuch as it neglects many factors such as α-adrenergic constriction. However, the integrative scheme is based on data that demonstrate longitudinal gradients for metabolic, myogenic, and flow-dependent responses [33]. The figure depicts the feed-forward sequences as gray arrows with Θ and the negative feedback loops as dark arrows denoted by Θ. If myocardial oxygen consumption were to suddenly increase during hemodynamic perturbations associated with exercise or excitement, there would be increased production of metabolites by the heart. In a normal heart an increase in metabolism produces vasodilation, which occurs preferentially in the smallest coronary arterioles [30,31]. Small coronary arterioles are also more sensitive to adenosine than are upstream vessels [11,33]. Metabolic dilation of these small arterioles would decrease upstream pressure. This would produce myogenic vasodilation of intermediate-sized arterioles, which possess greatest sensitivity to the effects of stretch. Myogenic and metabolic vasodilation would also cause a decrease in network resistance, allowing greater flow through the upstream larger arterioles and small arteries, thereby recruiting flow-dependent vasodilation of these vessels. The negative feedback loops are important to emphasize in this situation, because the upstream dilation allows for transmission of pressure to the downstream segments, which would brake further myogenic vasodilation (this is shown by the dark gray arrows denoted by Θ). Also, the overall decrease in resistance, and thus the increase in flow, would induce washout of the vasoactive metabolites and attenuate excessive metabolite-induced dilation (again shown by the negative feedback loop). Associated with the return of metabolic demands to baseline conditions, the opposite would happen to return coronary blood flow to basal levels. This scheme is predicated on normal functions of all segments, but in the situation of impaired endothelial flow-dependent vasodilation or exhaustion of metabolic vasodilator reserve that occurs with a severe stenosis, the proper adjustments cannot occur. Perhaps these inappropriate responses provide the basis for explaining the microvascular role in...
some coronary pathologies characterized by inadequate vasodilation to match myocardial metabolism.

8. Conclusion

Coronary blood flow is determined by coronary vascular resistance that is distributed throughout the vascular tree. However, the majority of coronary vascular resistance resides in the coronary microvasculature. The responses of vessels less than 150 μm in diameter to neural, hormonal, and mechanical stimuli are therefore of particular importance in understanding control of coronary blood flow. Development of new techniques to study the coronary microcirculation have now demonstrated that, even within the microcirculation, responses to the same vasoactive stimulus may differ between large and small arterioles. Therefore, studies of the mechanisms that contribute to determination of coronary vascular resistance must not treat the resistance vasculature as a single unit, but rather one must consider the heterogeneity of these mechanisms within the microcirculation. Consideration must also be given to venular responses. In vitro studies of isolated venules indicate that these vessels show active vasodilatory and vasoconstrictor responses to both physiological and pharmacological stimuli. Similarly, it cannot be presumed that endocardial and epicardial vessels display similar responses to vasoactive stimuli. The coronary microcirculation must be studied as a conglomerate of distinct parts, each of which contributes to overall control of pressure and flow.

Coronary blood flow must be closely regulated in order to meet the constantly changing metabolic demands of the myocardium. This requires rapid changes in coronary vascular tone over a wide range. Such precise, yet extensive regulation can only be achieved by integration of a variety of control mechanisms. It has become clear that no single mechanism predominates in control of coronary vascular tone: neural, hormonal, and local control mechanisms all participate. A lack of balance between these mechanisms is often apparent in disease states which are characterized by inadequate control of coronary blood flow. In addition to a balance between control mechanisms, close control of coronary blood flow is facilitated by heterogeneous distribution of these mechanisms within the coronary circulation. Coronary vascular tone is not determined by global responses of the entire coronary bed to vasoactive stimuli, but rather by coordination of diverse responses within local segments of the coronary circulation.

References


