Profound Spatial Heterogeneity of Coronary Reserve
Discordance Between Patterns of Resting and Maximal Myocardial Blood Flow


We examined the ability of individual regions of the canine left ventricle to increase blood flow relative to baseline rates of perfusion. Regional coronary flow was measured by injecting radioactive microspheres over 90 seconds in seven anesthetized mongrel dogs. Preliminary experiments demonstrated a correlation between the regional distributions of blood flow during asphyxia and pharmacological vasodilatation with adenosine (mean r=0.75; 192 regions in each of two dogs), both of which resulted in increased coronary flow. Subsequent experiments, during which coronary perfusion pressure was held constant at 80 mm Hg, examined the pattern of blood flow in 384 regions (mean weight, 106 mg) of the left ventricular free wall during resting flow and during maximal coronary flow effected by intracoronary adenosine infusion. We found that resting and maximal flow patterns were completely uncorrelated to each other in a given dog (mean r=0.06, p=NS; n=3 dogs). Furthermore, regional coronary reserve, defined as the ratio of maximal to resting flow, ranged from 1.75 (i.e., resting flow was 57% of maximum) to 21.9 (resting flow was 4.5% of maximum). Thus, coronary reserve is spatially heterogeneous and determined by two distinct perfusion patterns: the resting (control) pattern and the maximal perfusion pattern. Normal hearts, therefore, contain small regions that may be relatively more vulnerable to ischemia. This may explain the patchy nature of infarction with hypoxia and at reduced perfusion pressures as well as the difficulty of using global parameters to predict regional ischemia. Despite the wide dispersion of coronary reserve, we found, by autocorrelation analysis, that reserve in neighboring regions (even when separated by a distance of several tissue samples) was significantly correlated. This also applied to patterns of resting myocardial flow. Thus, both resting coronary blood flow and reserve appear to be locally continuous and may define functional zones of vascular control and vulnerability, respectively. (Circulation Research 1990;67:319–331)

It is now well accepted that myocardial perfusion is spatially heterogeneous. Not only is this lack of uniformity apparent when one considers blood flow to the heart by layer (i.e., a given depth from the epicardial surface) but also when one examines coronary flow to regions within a layer. Regional variability of perfusion, which has been observed in the hearts of all species studied thus far, is quantitatively much larger than the error of the techniques used to measure blood flow. For example, have found that even after correcting for methodological error and temporal variability, flow to small regions of the left ventricle of conscious baboons ranges almost sixfold in its extremes. Although the basis for such heterogeneity remains to be explained, differences in local metabolic needs, perhaps secondary to differences in regional function, have been suggested. Physiologically more interesting than resting differences in perfusion, however, is the ability of individual regions to meet blood flow demands in times of need (e.g., with exercise or decreased perfusion pressure). Studies of regional blood flow in the absence of coronary tone demonstrate that the distribution of maximal flow—as with resting flow—is extremely heterogeneous. This suggests that seem-
ingly intrinsic properties, such as maximal regional blood flow, vary significantly throughout the heart.

In this study, we examined the relative ability of myocardial regions to increase blood flow by comparing resting and maximal flows in individual small regions of the heart. We wondered whether regions with higher than average resting flow also have higher than average maximal flow. The correspondence between resting and maximal perfusion patterns determines the spatial distribution of coronary reserve. Poor correlation between these patterns would result in a wide range of regional reserve, whereas a good match implies a fairly uniform ability to increase flow.

To evaluate these possibilities, we studied the distribution of left main coronary artery (LMCA) blood flow using separately labeled sets of radioactive microspheres to compare resting and maximal regional blood flow at normal perfusion pressures. Initial experiments addressed qualitative aspects of this comparison, as well as the relation between pharmacological manipulation of vascular tone and the physiological challenge of asphyxia. In the remaining experiments, coronary perfusion pressure was held constant to allow absolute comparisons of regional blood.

**Materials and Methods**

**Experimental Preparation**

We studied seven mongrel dogs of both sexes weighing 23.9–29.5 kg. Each was sedated with sodium thiopental (25 mg/kg i.v.) before endotracheal intubation and mechanical ventilation with room air supplemented by oxygen at tidal volumes of 15–20 ml/kg. Anesthesia was maintained with 1% halothane. Periodic arterial blood gas measurements allowed us to adjust ventilatory settings to keep pH, PO2, and PCO2 within normal limits.

Femoral arterial catheters were placed for blood gas sampling, aortic pressure monitoring, and microsphere reference samples. After left thoracotomy via the fifth intercostal space, the heart was suspended in a pericardial cradle. In two dogs, catheters were inserted into the left atrium via the left atrial appendage to inject microspheres and adenosine.

In the five remaining dogs, LMCA perfusion pressure was held constant with a pressurized femoral-coronary shunt under servo control. After anticoagulation with heparin (10,000 units i.v. and then 5,000 units hourly), a Gregg cannula, as described by Sarnoff et al., was inserted through the left subclavian artery, placed just inside the left coronary ostium, and secured with an external silk ligature around the proximal LMCA. Pressure at the distal tip of the cannula was digitally sampled at 20 Hz and compared with a previously calibrated target pressure. This feedback was used to modulate the opening and closing of an air valve connecting a 250–mm Hg pressure source to a sealed air-blood level in the femoral-coronary circuit (Figure 1). The servo system ensured that mean coronary pressure was maintained within 0.5% of target pressure.

**Blood Flow Measurements With Radiolabeled Microspheres**

Regional coronary blood flow was measured using the radioactive microsphere technique. Each dog received either intracoronary or left atrial injections of 3–10 million 15±1.5-μm diameter (mean±SD) microspheres over 90 seconds. These spheres contained the gamma-emitting nuclides 153Gd, 57Co, 114mIn, 51Cr, 113Sn, 85Sr, 99Nb, 54Mn, and 65Zn (3M, St. Paul, Minn.; New England Nuclear, Boston, Mass.), each corresponding to a particular coronary blood flow determination. To prevent aggregation, the spheres were suspended in saline with surfactant (0.01% Tween 80). In the dogs receiving left atrial injections, femoral blood was collected continuously during and for 2 minutes after the end of the injection. The timed femoral artery withdrawal provided us with a microsphere-containing reference sample collected at a known flow rate. This allowed us to calculate absolute blood flow in a tissue sample using the following relation:

\[
Q_{\text{sample}} = Q_{\text{reference}} \times \frac{C_{\text{sample}}}{C_{\text{reference}}}
\]
where $Q_{\text{sample}}$ is blood flow in the sample, $Q_{\text{reference}}$ is the rate of reference flow, $C_{\text{sample}}$ is the radioactivity of the tissue sample, and $C_{\text{reference}}$ is the radioactivity of the reference sample.

Regional myocardial blood flow measurements based on intracoronary microsphere injections were referenced to total LMCA flow as determined with an in-line electromagnetic flow transducer. The above relation holds if $Q_{\text{reference}}=\text{LMCA flow}$ and $C_{\text{reference}}=\text{total activity of the heart}.$

After the final microsphere injection, each dog was killed with a large dose of pentobarbital, and its heart was removed. The left ventricular free wall was trimmed into a square approximately 6 cm on a side, fixed in formaldehyde, and subsequently cut into subendocardial, middle, and subepicardial layers. Each layer was further subdivided into 128 regions (Figure 2) with a mean piece weight of 106±30 mg (mean±SD); in the dogs receiving left atrial microsphere injections, there were $3 \times 64=192$ regions (mean weight, 142±44 mg). The radioactivity of each of these samples was measured with a gamma detector. In the dogs receiving intracoronary injections, the remainder of the heart was homogenized and counted so that total heart radioactivity could be calculated. In all, there were a total of 6,912 measurements in 2,304 regions of seven hearts.

The radioactivity of each sample of myocardium was determined using a NaI(Tl) detector (Tracer Analytic, TM Analytic, Brandon, Fla.) and a multichannel pulse height analyzer (Norland Corp., Fort Atkinson, Wis.), along with a NOVA-3 minicomputer (Data General, Southboro, Mass.). Individual nuclide activities were determined using the least-squares method described by Baer et al. All samples were counted for 3 minutes. The measured activity of each tissue sample was corrected for differences in geometric configuration by applying an empirically established relation (least-squares linear fit) between sample weight and the measurement of known activities. All blood samples were the same height and were counted using calibrations based on reference samples placed on the bottom of sample vials. We estimated the errors of counting inherent to the NaI(Tl) detector by using the goodness of fit of nuclide separation to approximate the variance in the determinations of the activity of each individual nuclide.

**Experimental Protocol**

The initial phase of the study focused on the patterns of coronary perfusion during increased coronary flow due to intra-atrial adenosine administration (350 μg/kg/min) or asphyxia induced by disconnecting the respirator for 90 seconds. Control measurements were obtained before and between interventions in the two dogs studied in this part of the protocol. In the first dog, asphyxia preceded adenosine infusion. This order was reversed in the second dog. In both dogs regional blood flow was measured using left atrial injections of microspheres.

For the remainder of the study, coronary perfusion pressure was maintained at a pressure of 80 mm Hg during all conditions in all dogs. In five dogs, we examined the resting pattern of myocardial perfusion. In three of these, regional blood flow was also determined during maximal coronary vasodilatation with intracoronary infusions of adenosine (20 μg/kg/min). Maximal coronary vasodilatation was confirmed in these dogs by temporarily doubling the infusion rate without evidence of a concomitant increase in left coronary blood flow.

In some of the dogs, temporal variability of regional blood flow and experimental error of the microsphere technique were evaluated by using sequential or simultaneous microsphere injections containing different radionuclides.

**Numerical Methods**

Statistical comparisons of coefficients of variation (the ratio of the standard deviation of a distribution of measurements to its mean) were performed using the technique of Lewontin, as described by Zar. Correlation coefficients ($r$) were calculated in a standard fashion and refer to a "simple" or Pearson product-moment correlation coefficient. Statistical comparisons of correlation coefficients were made using standard techniques. Comparisons among layers were made by two-way analysis of variance.

To assess local continuity, we studied autocorrelations of flow and reserve within a layer with respect to distance. The autocorrelations were calculated by evaluating the correlation of flow (or reserve) in myocardial regions a given distance apart, without regard to direction (i.e., isotropically). Specifically, we first estimated covariance for each distance ($d$) using the following relation:
TABLE 1. Assessing Components of Error in the Microsphere Technique

<table>
<thead>
<tr>
<th></th>
<th>Dogs 1 and 2</th>
<th>Dogs 3–7</th>
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<tbody>
<tr>
<td>Number of spheres/region</td>
<td>624</td>
<td>1,103</td>
</tr>
<tr>
<td>Counts (3 min)</td>
<td>14,069</td>
<td>25,457</td>
</tr>
<tr>
<td>Separation error (cpm)</td>
<td>605</td>
<td>517</td>
</tr>
<tr>
<td>Mean combined</td>
<td>...</td>
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Dogs 1 and 2, dogs in initial phase of study with increased coronary flow due to intra-atrial adenosine administration (350 μg/kg/min); dogs 3–7, dogs with coronary perfusion pressure maintained at 80 mm Hg.

\[ V(d) = \frac{1}{n_d} \sum_{\text{all regions } a,b \text{ distance } d \text{ apart}} [Q_a - \overline{Q}][Q_b - \overline{Q}] \]

where \( V(d) \) is the covariance for distance \( d \), \( n_d \) is the number of regions distance \( d \) apart, \( \overline{Q} \) is mean flow per layer, and \( Q_a \) and \( Q_b \) are flows in regions \( a \) and \( b \), respectively, that are distance \( d \) apart. The correlation function \( r(d) \) is the ratio of the spatial covariance to the variance of flow within a layer \( [V(0)] \); that is,

\[ r(d) = \frac{V(d)}{V(0)} \]

We estimated the significance (versus zero) of an autocorrelation for a specific distance by comparing each point to \( 2/\sqrt{n_d} \); this is a commonly employed approximation to twice the standard deviation (personal communication, D.R. Brillinger, University of California, Berkeley).

Assessment of the Microsphere Method

The errors in microsphere blood flow measurements have, in part, a statistical basis.14–17 This is a result of the discrete and, therefore, stochastic nature of both microsphere entrapment and radioactive disintegrations. The following relation has been shown to account for all of the variability in relative measurements and the majority of variability in absolute blood flow measurements with radioactive microspheres14:

\[ \text{RE}_{\text{total}}^2 = \text{RE}_{\text{counting}}^2 + \text{RE}_{\text{spheres}}^2 \]

where \( \text{RE}_{\text{total}} \) is the total relative error of a microsphere measurement, \( \text{RE}_{\text{counting}} \) is the error of separation and radioactive counting, and \( \text{RE}_{\text{spheres}} \) is the Poisson error associated with the number of spheres in a sample. Traditionally, only the latter component, \( \text{RE}_{\text{spheres}} \), has been considered when estimating error.15 In dogs requiring microsphere reference samples, additional error must be included.14,16

Based on the above relation, a theoretical estimate14 of the mean error of coronary blood flow due to the microsphere technique can be calculated for both parts of the study (Table 1). Simultaneous injections of separately labeled microspheres during some of the

**Figure 3.** Plots showing empiric assessment of the microsphere technique using simultaneous injections. Microspheres containing different isotopes were injected during different manipulations of coronary flow.
coronary blood flow manipulations in the initial dogs allowed us to evaluate the error of the microsphere method empirically (Figure 3). Because of the lower coronary concentrations of microspheres in heart tissue with left atrial injections, blood flow determinations in the initial dogs had significantly more error than in the remaining dogs. It should be noted that examinations of the variability of microsphere measurements of regional blood flow only assess the reproducibility of the technique and do not encompass systematic errors of the microsphere technique.9

Results

Initial Regional Coronary Flow Measurements

Table 2 outlines the order of interventions, the mean LMCA flow, and the aortic blood pressure in the dogs receiving left atrial microsphere injections. During intra-atrial adenosine administration, mean blood pressure fell in both dogs but was increased markedly after 90 seconds of asphyxia.

Figure 4 demonstrates the distribution of regional coronary blood flow for these dogs. Although mean LMCA flow was greater during asphyxia than during adenosine infusion, coronary vascular resistance, defined as the ratio of diastolic aortic pressure to mean coronary flow, was lower during adenosine infusion (Table 2). Figure 5 depicts a comparison of coronary flow in regions of the left ventricular free wall during different experimental conditions: control versus control, adenosine versus asphyxia, and adenosine versus control. The correlation was significant only when we compared either 1) regional flow during adenosine administration to flow during asphyxia or 2) repeat measurements of resting regional flow (p<0.001). The distribution of coronary flow was poorly, or not significantly, correlated when we compared resting regional flow (during control conditions) with flow during either high coronary flow condition (i.e., asphyxia or adenosine).

Constant Coronary Perfusion Pressure

To eliminate the effect of changes in aortic pressure on coronary pressure during manipulations, coronary perfusion pressure was held constant at 80 mm Hg via a Gregg circuit.10 Direct access to the coronary circulation also provided a way to adminis-

![FIGURE 4. Histograms showing distribution of regional coronary flow during different experimental conditions in dogs receiving left atrial microsphere injections. Arrows indicate mean flow for each condition.](image)
ter adenosine locally and, therefore, reduce total systemic dose (intracoronary doses were 1/17 of intra-atrial doses).

Regional Distribution of Coronary Blood Flow

We assessed the distribution of regional blood flow in five dogs with intracoronary microsphere injections. Figure 6 illustrates a typical distribution from a single dog. This dog had a coefficient of variation of regional flows of 17.6%. Although mean flow was 1.58 ml/g/min, the extremes of flow ranged more than threefold. The mean coefficient of variation in all dogs was 24.3% (n=5). We also examined regional flow by layer and found no consistent pattern among dogs (Table 3).

Using intracoronary infusions of adenosine, we examined the regional pattern of coronary perfusion in the absence of vascular tone. Figure 6 displays the distribution of maximal regional flow from a single dog. Although mean flow rose to 8.14 ml/g/min, the distribution of regional flow ranged more than fourfold. The coefficient of variation was 33.1% in this dog and averaged 30.4% for all three dogs (significantly different from the resting flow coefficient of variation, p<0.05). When we assessed flow by layer (Table 3), we found that maximal flow was significantly less in the subepicardial layer than in the inner layers (p<0.05 by two-way analysis of variance).

Temporal Stability of Flow Distributions

The stability of these distributions was assessed by injecting a second label 5 minutes after the first (Figure 7). We found strong correlation between repeat measurements of resting flow in dogs thus

**FIGURE 5.** Plots showing comparisons of regional coronary flow during different experimental manipulations in dog 1 (top panels) and dog 2 (bottom panels). Not depicted are plots of asphyxia versus control, which had correlation coefficients in dog 1 and dog 2 of 0.38 and 0.21, respectively.

**FIGURE 6.** Histograms showing distribution of regional coronary flow in a single dog receiving intracoronary microsphere injections. Left panel: Resting (control) distribution. Right panel: Distribution of maximal regional flow for the same dog.
studied (mean $r=0.93$, $n=3$). In other words, regions that tended to have low flows continued, over time, to have low flows, and regions with higher than average blood flow remained high. The patterns of blood flow during maximal coronary vasodilatation were also found to be very stable over time (Figure 7).

**Regional Coronary Reserve**

Because resting and maximal regional blood flow displayed heterogeneous distributions, we compared flow in each region during resting flow with the region's blood flow during pharmacological coronary vasodilatation. Figure 8 demonstrates the lack of correlation between resting and maximal flow patterns in all dogs examined (mean $r=0.06$, $p=NS$). Pieces with high resting flow, therefore, had no greater tendency to have above-average maximal flow than pieces with low resting flow.

Figure 9 contains histograms of the distribution of regional coronary flow reserve for these dogs. We defined reserve as the ratio of maximum flow to resting flow at a given perfusion pressure. Note that although the mean reserve in these dogs was 6.2, it ranged from 1.75 (i.e., resting flow was 57% of maximum) to 21.9 (resting flow was 4.5% of maximum). In addition, distributions were skewed to the right, suggesting a physiological lower limit for coronary reserve.

**Continuity of Flow and Flow Reserve Patterns**

Finally, we examined the degree of local continuity of flow and flow reserve using autocorrelation, that is, by comparing all possible pairings of pieces within a layer (there were 8,128 such comparisons per layer per dog) and calculating the mean correlation among flows of regions a given distance apart (Figure 10). This provided a way to compare relative flows in neighboring regions and to demonstrate that, although regional flow (and reserve) may be highly variable within a layer, it appears to be locally continuous since flow correlation consistently increased at shorter and shorter distances. One can assess the size of these "zones of continuity" by estimating $r_{1/2}$, the half-correlation distance (i.e., the distance at which the autocorrelation falls to 0.5). For simplicity, we estimated $r_{1/2}$ from the best-fit line predicted by the least-squares method. The mean $r_{1/2}$ for all layers during control conditions was 3.5 mm ($n=15$ layers in five dogs). The mean $r_{1/2}$ of the maximum flow patterns was 8.4 mm, and the mean $r_{1/2}$ of regional reserve was 7.6 mm ($n=9$ layers in three dogs). We noted that the half-correlation distances for reserve were generally longer in the subepicardium than in the subendocardium, suggesting greater continuity of regional reserve in the outer layers of the heart (Figure 11). Figure 12, a perspective plot from a single dog, graphically depicts the increasing continuity of regional reserve in the direction of the subepicardium.

Another way to assess local continuity is to note the maximum distance at which there is significant autocorrelation. Although this parameter is more sensitive to the errors of the microsphere technique (i.e., increased methodological error would shorten the maximum distance of correlation), it sets a minimum on the range over which flow in two regions is correlated. The mean maximum distance of significant correlation was 9.2±3.1 mm for resting flow ($n=15$ layers). It was 12.0±4.4 and 11.4±3.5 mm for maximum flow and regional reserve, respectively ($n=9$ layers).

**Table 3. Mean Regional Flow and Reserve by Ventricular Layer**

<table>
<thead>
<tr>
<th>Dog</th>
<th>Endo</th>
<th>Mid</th>
<th>Epi</th>
<th>Endo</th>
<th>Mid</th>
<th>Epi</th>
<th>Endo</th>
<th>Mid</th>
<th>Epi</th>
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<tr>
<td>3</td>
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<td>1.28</td>
<td>0.94</td>
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<td></td>
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<tr>
<td>4</td>
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<td>1.13</td>
<td>1.26</td>
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<td>10.1</td>
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<td></td>
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<tr>
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<td>1.00</td>
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<tr>
<td>7</td>
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<td>1.02</td>
<td>1.01</td>
<td>9.45</td>
<td>9.46</td>
<td>6.91</td>
<td>8.92</td>
<td>9.27</td>
<td>6.84</td>
</tr>
<tr>
<td>Mean</td>
<td>1.19</td>
<td>1.11</td>
<td>1.12</td>
<td>7.86</td>
<td>7.72</td>
<td>5.60*</td>
<td>6.63</td>
<td>6.95</td>
<td>5.00*</td>
</tr>
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</table>

*Significantly different from the inner layers by two-way analysis of variance ($p<0.05$ and $p<0.01$ for maximum flow and reserve, respectively).

**Figure 7. Plots evaluating the temporal stability of the distribution of regional blood flow. A second set of microspheres containing a different nuclide was injected 5 minutes after the first. Left panel: Comparison of repeated measurements of regional control flow. Right panel: Comparison of repeated measurements of maximal regional flow. (This dog is also depicted in Figure 6).**
We also analyzed the local correlation of resting flow and reserve in the radial direction (i.e., layer versus layer). We compared regional flow and reserve in corresponding regions in the subendocardial, middle, and subepicardial layers. Mean correlation (r) between adjacent layers was 0.63 and 0.67 for resting flow and reserve, respectively (p<0.001). Mean correlation of flow and reserve between corresponding pieces in the subendocardium and subepicardium was 0.42 and 0.48, respectively (p<0.01).

**Discussion**

**Critique of Method**

As our assessment of regional flow is based solely on microsphere measurements, the validity of this

**FIGURE 8.** Plots comparing regional blood flow at rest with regional flow during maximal pharmacological vasodilatation with adenosine in three dogs. (The dog depicted on the upper left is in Figures 6 and 7.)

**FIGURE 9.** Histograms showing distribution of coronary reserve in three dogs at a perfusion pressure of 80 mm Hg. Coronary reserve is defined as the ratio of maximal regional blood flow to control flow. Note the consistent rightward skew.
technique is of primary importance in the interpretation of our results. The accuracy of the radioactive microsphere technique has been examined extensive-
ly,7-9,11,15,16 and recently our laboratory has also studied this question in detail.14 There are two major concerns with the microsphere technique: the degree to which flow is proportional to sphere deposition and the effect of microspheres on the microcirculation.

We evaluated the reproducibility of measurements with microspheres both theoretically (Table 2) and empirically (Figure 3), using simultaneous injections of microspheres containing different isotopes. These estimates of the error in flow (i.e., the stochastic errors14) were minimal when compared with measured regional heterogeneity. Another potential source of error with the microsphere technique is the possibility that the rheological properties of the spheres may prevent them from reflecting true blood flow.8,9 Although large microspheres (e.g., 50 μm in diameter) have been demonstrated to deposit preferentially in areas of high flow,7-9 this appears to be less important with smaller spheres such as the 15-μm spheres used in this study. Fifteen-micron spheres have been shown to correlate well with nonparticulate tracers, including iododesmethylimipramine.8,9 Thus, although it is likely that there are significant systematic, as well as random, errors associated with the microsphere technique, they appear to be much smaller than the observed heterogeneity of flow and flow reserve we have found in this study. Therefore, it is unlikely that our findings are qualitatively affected by these errors.

Another concern with the microsphere technique has been the degree of circulatory impairment after a microsphere measurement. Surprisingly, even with large numbers of spheres, the amount of microvasculature at risk with most measurements is small. In our study, dogs that had four coronary microsphere injections received a total of approximately 20 million microspheres. We estimate this number of spheres to affect only about 4% of capillaries.8 The apparent temporal stability of repeat measurements of regional flow (Figures 5 and 7) also suggests that progressive circulatory impairment was minimal.

Another consideration is that our system of perfusing the LMCA may have resulted in a heretofore unappreciated effect on measured heterogeneity. Data from the initial dogs, which agrees qualitatively with those from dogs in which the LMCA was cannulated, suggest that our findings were not due to an artifact associated with experimental control of coronary perfusion. Finally, it is possible that the use of anesthetized, open-chest dogs affected our measurements of flow and regional reserve. Previous studies2,3,5-9,19 in conscious and anesthetized ani-
mals, though, have demonstrated similar magnitudes of flow heterogeneity. The effect of volatile anesthesia on regional flow in the absence of coronary tone has been less well studied.

Regional Coronary Flow and Reserve

In our study we examined the regional distribution of LMCA perfusion in the dog, specifically, the pattern of blood supply to the left ventricular free wall. We focused on the left ventricular free wall because of its exclusive LMCA supply. In the initial experiments we attempted to assess, primarily in a qualitative way, the regional distribution of blood flow during baseline conditions and two fundamentally different manipulations, both of which resulted in marked increases in total coronary flow. Because the number of microspheres per tissue sample was limited in the dogs with left atrial microsphere administration, there was a moderate amount of error (Table 1). In addition, the opposing systemic effects of these manipulations on aortic, and thus coronary, perfusion pressure prevented absolute comparisons of regional blood flow during different experimental conditions. Nevertheless, comparisons of relative regional flow provided several interesting observations, some of which are corroborated by findings in the latter part of this study.

First, although total LMCA flow varied with time, the relative distribution of resting regional flow was stable, even when compared before and several minutes after an experimental manipulation (Figure 5). Second, there were strong correlations between patterns of regional perfusion when coronary flow was increased, although the methods used to increase coronary flow were quite different and actually had opposite effects on arterial blood pressure (Table 2). This is of interest because several laboratories have recently demonstrated that, even during significant reductions in coronary flow, vasodilatory reserve is not exhausted.20,21 Our experiments demonstrate that although the absolute change in regional vascular resistance during adenosine administration was larger than with asphyxia, the relative change in a given region was approximately the same. This suggests that the pharmacological recruitment of reserve qualitatively, if not quantitatively, mimics the physiological response to increased blood flow demand during hypoxia and hypercapnia.

Finally, there was little correlation between perfusion patterns when comparing relative regional flow at rest and during increased coronary flow. It is important to note that in the preliminary experiments we did not directly measure coronary flow and, since our experimental manipulations were limited by their systemic effects, we did not test whether coronary flow was maximal.

To overcome these limitations, we controlled coronary blood flow in the remainder of the experiments by means of a Gregg circuit.10 This had several advantages, the most important of which was control of LMCA perfusion pressure. Because in the absence of coronary autoregulation regional blood flow depends on perfusion pressure, accurate comparisons of regional flow require constant coronary pressure. In addition, direct access to the coronary circulation enabled us to use doses of adenosine that minimized systemic effects and thus to avoid significant changes in afterload that had been observed to alter transmural distribution of regional blood flow.22 Direct access also ensured that the number of microspheres trapped by the myocardium would be independent of the type of experimental manipulation and its effect on the distribution of cardiac output.

As in the initial dogs, we found significant spatial heterogeneity of both baseline and maximum regional flow. This agrees with several previous studies.1-9,19 In addition, these distributions of flow were found to be relatively constant over time (Figure 7). Studies in conscious animals demonstrated that regional perfusion patterns can be quite stable over several hours. Despite the stability of each flow distribution, however, there was no correlation between resting and maximal regional flow patterns (Figure 8). Thus two distinct perfusion patterns determine coronary flow reserve: the resting (or control) pattern and the maximal flow pattern.

The resting distribution of regional myocardial blood flow is of particular interest, because there is evidence that under most conditions regional perfusion closely reflects local metabolic needs. First, it is well known that the resting coronary arteriovenous oxygen difference is near maximal (approximately
60–70%) and does not appreciably widen with exercise or reductions in coronary flow.23,24 This suggests that myocardial blood supply and demand are optimally matched because little oxygen, even at rest, passes unextracted through the myocardium. Second, changes in regional contractility following changes in coronary blood flow also suggest that regional perfusion is coupled to metabolic demand.25–27 Finally, the increased myocardial oxygen requirement during high coronary flow, a phenomenon known as the Gregg effect,28 also provides an “incentive” for tight control of coronary flow. These points suggest that the heterogeneous distribution of regional coronary flow probably reflects an equally heterogeneous metabolic demand.

The distribution of maximal regional blood flow, on the other hand, appears to reflect factors that are largely mechanical and quite separate from those that influence regional perfusion when vascular tone is intact. Coronary driving pressure, length of distal, and extraluminal tissue pressure are major determinants of maximum regional flow.29 Ultimately though, the underlying vascular architecture establishes the upper limit of the maximum blood flow possible to a given region of myocardium. Studies of the regional vascularity of the heart reveal arteriolar and capillary networks that have random branching patterns.30,31 Indeed, recent models of regional blood flow now incorporate the stochastic nature of the vascular tree.32,33 Thus, a combination of factors influences regional flow in the absence of vascular tone and results in the spatially heterogeneous maximal perfusion pattern found in this and previous studies.6 Because resting and maximal regional blood flow patterns appear to be determined by distinct factors, there is little reason to expect them to be related.

The apparent mismatch between perfusion patterns has several implications. First, it implies there are markedly different amounts of reserve from one region to another. In this study, we had tissue samples with resting blood flows as low as 4.5%, and as high as 57%, of maximal blood flow in the region (Figures 8 and 9). Thus, especially when one considers that in humans mean coronary blood flow can increase more than fourfold during heavy exercise,24,34 some regions required resting rates of perfusion that were relatively close to maximal capacity.

Extreme spatial heterogeneity of reserve may explain the patchy nature of myocardial infarction after hypoxia or global reductions in coronary perfusion pressure, for example, with cardiac tamponade, shock, aortic valvular stenosis, or severe coronary stenosis.35 Regions with low coronary flow reserve simply may be more vulnerable. Studies using an ultraviolet fluorescence technique to measure reduced nicotinamide adenine dinucleotide have demonstrated small regions of ischemia on the epicardial surface of perfused rat hearts either with low perfusion pressure or during hypoxia.36,37 This suggests that the supply of oxygen can become locally insufficient. Other tissues, for the same reasons, may contain multiple foci of vulnerability. After decreases in renal perfusion, for instance, ischemic damage to the kidney is often patchy.38

Although the distribution of regional reserve may explain the focal nature of infarction in the setting of reduced perfusion pressure, it does not explain the frequently subendocardial distribution of these patchy infarcts. In fact, when we examined reserve by layer, we found slightly less reserve at 80 mm Hg in the subepicardium than in the middle and inner layers (Table 3). Several investigators27,39 have demonstrated, however, that as coronary perfusion pressure is lowered, mean reserve falls more rapidly in the inner layers. Thus, other factors, such as transmural tissue pressure or perhaps the movement of blood flow from inner to outer layers during systole,40–43 may significantly compromise subendocardial reserve at low perfusion pressures. Our study suggests, though, that even when global factors (e.g., elevated diastolic ventricular pressure or reduced perfusion pressure) place the subendocardium at risk for ischemia, there will be islands of relatively greater vulnerability within the inner layers of the heart.

The spatial heterogeneity of reserve may also explain the difficulty in predicting regional ischemia using global measurements that do not take into account the wide distribution of regional reserve. For instance, it may be that some patients with symptoms of stress-induced angina despite normal coronary anatomy (as assessed by angiography)44,45 have regionally inadequate coronary reserve. Conversely, regional variability of reserve may make it misleading to extrapolate about the heart as a whole using a limited number of regional measurements46 (e.g., biopsies and regional pH measurements). In this study, we found that, on average, measurements of regional reserve were completely unrelated (i.e., not significantly correlated) at distances greater than 11.4 mm. Thus, it may be wise to use several spatially distinct samples in regional assessments of ischemia.

Our method of regional flow analysis allowed us to examine a final consideration: the local continuity of regional perfusion and reserve within layers of the heart. This issue is separate from the assessment of regional blood flow dispersion (e.g., the coefficient of variation), which does not take into account the degree of local variation among neighboring regions. For example, when a layer of myocardium has greater blood flow in the apex than the base, it necessarily has a dispersion about the mean flow of the layer. If, in the same layer, regional flow gradually, but steadily, changed from apex to base, total dispersion could remain high while local discontinuity (reflecting only the change from piece to piece in the direction of the base-apex axis) would be minimal. In fact, we had several instances in which the coefficient of variation of regional flow increased or stayed the same from one layer to the next, yet local discontinuity fell (i.e., half-correlation distance, r1/2, increased; Fig-
ure 12). Of note, in all dogs in which we measured reserve, we consistently found smaller half-correlation distances in the inner layers, implying that regional reserve was less continuous in the subendocardium and midwall than in the subepicardium (Figures 11 and 12). In all dogs, we observed significant correlation of flow among neighboring regions of a layer. The correlation was often significant over a distance of several regions and consistently improved at shorter distances (Figure 11). Bassingthwaighte et al.19 have recently proposed a fractal model of regional coronary flow that predicts a moderately strong correlation between local flows. In the current study, we have used autocorrelation to directly demonstrate the degree of local flow continuity.

Because flow in adjacent myocardial regions is correlated, these areas may be considered to be contained within a functional zone of vascular control. Using \( r_{1/2} \) as a conservative estimate of the radius of such a zone, we calculate a mean size of 0.38 cm². Such a unit encompasses several hundred arterioles.47,48 Similarly, when examining regional reserve, the \( r_{1/2} \) defines a "zone of vulnerability," which, in our study, had a mean size in the dog of approximately 1.8 cm². Because reserve appeared to be less continuous in the subendocardium than the subepicardium, functional areas of reserve are probably smaller in inner layers of the heart. The mechanisms contributing to continuity of flow or flow reserve in regions much larger than the area perfused by a single arteriole—the major locus of control of vascular resistance49—are unknown. Perhaps macroscopic factors, such as intraventricular or transmural pressure, affect the caliber of vessels in a stochastic microvasculature (and thus the maximal perfusion pattern) and influence local metabolic needs (and thus the resting perfusion pattern) in a continuous way.

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References


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