Transitional Blood Flow Zones between Ischemic and Nonischemic Myocardium in the Awake Dog

Analysis Based on Distribution of the Intramural Vasculature

Robert H. Murdock, Jr., David M. Harlan, James J. Morris, III, William W. Pryor, Jr., and Frederick R. Cobb

SUMMARY. The present study evaluates the transitional or border zone of intermediate blood flow reduction between nonischemic and ischemic regions after acute coronary artery occlusion in chronically instrumented dogs, using methods that minimize an admixture of ischemic and nonischemic myocardium in the tissue analyzed. The regions perfused by occluded and nonoccluded vessels were identified by tracing the extra and intramural distribution of the coronary vasculature from postmortem angiograms. Regional blood flow was evaluated in serial 3-mm-wide epicardial and endocardial zones from outside and inside the interface between occluded and nonoccluded vessels. The zone of intermediate reduction in blood flow between nonischemic and ischemic regions occurred in the first 3-mm section immediately inside the region supplied by the occluded vasculature. Mean blood flow in this region was reduced to 58 ± 6% and 61 ± 5% (±SEM) of nonischemic region blood flow at the lateral and medial epicardial margins, respectively, and 47 ± 5% and 45 ± 6% at the lateral and medial endocardial margins, respectively. In the remaining ischemic zone, significant differences in blood flow existed between epicardial and endocardial layers; these differences were highly variable between animals. The data indicate that when the analysis of regional blood flow following acute ischemia is based on the anatomic distribution of the coronary vasculature, the transitional or border zone of intermediate reduction in blood flow is limited to a narrow zone immediately inside the occluded vasculature. Studies performed in acutely anesthetized dogs in which the occluded region was perfused via a two-chamber blood reservoir that allowed maintenance of perfusion and exclusion of microspheres from the circumflex region indicate that intermediate reductions in blood flow at the border of the ischemic zone resulted from an admixture of normal myocardium and, thus, do not represent a border zone of intermediate ischemia. (Circ Res 52: 451-459, 1983)

Following proximal occlusion of a major coronary artery in the canine model, the fraction of the left ventricle that becomes ischemic, the regional distribution of blood flow to the ischemic region, and the amount of myocardium that eventually becomes infarcted are highly variable (Becker et al., 1973; Rivas et al., 1976; Marcus et al., 1975; Jugdutt et al., 1979a; Swain et al., 1980). Major determinants of the extent of ischemic injury and infarction include the dimensions of the ischemic or risk region (Jugdutt et al., 1979b; Reimer and Jennings, 1979; Factor et al., 1981) and blood flow to the ischemic myocardium (Rivas et al., 1976; Jugdutt et al., 1979b; Cobb et al., 1982).

Considerable controversy exists concerning the transitional or border zone between ischemic and nonischemic zones. One view is that the ischemic region consists of a central zone of greatest ischemia that is surrounded by significant transitional zones characterized by intermediate reductions in blood flow (Becker et al., 1973; Jugdutt et al., 1979a, 1979b; Vokonas et al., 1978), intermediate myocardial injury, and incomplete necrosis (Vokonas et al., 1978; Cox et al., 1968; Hearse et al., 1977; Kjekshus and Sobel, 1970; Maroko et al., 1971, 1977; Braunwald et al., 1974; Sobel and Shell, 1973). According to this concept, transitional zones of intermediate ischemia result from preferential perfusion via the nonoccluded vasculature that borders the ischemic region. Other investigators have concluded that the transition from nonischemic to ischemic tissue is sharp and that a significant transitional or border zone of intermediate ischemia or myocardial injury does not exist (Reimer and Jennings, 1979; Factor et al., 1978; Hirzel et al., 1977; Factor et al., 1981; Yellon et al., 1981). The latter studies suggest that an apparent rather than a real border zone of intermediate ischemia and myocardial injury or necrosis may result from techniques used for tissue sampling and analysis and/or the anatomic arrangement of the microcirculation. A transitional zone of ischemia and/or tissue necrosis may result if analyses are performed on tissue samples that contain an admixture of tissue even though the interface between ischemic and nonischemic tissue is sharp.

The dimension of a transitional or border zone of ischemia and ischemic injury is pertinent to the concept of intervention therapy. It may be reasoned that a large zone of intermediate ischemia will increase the potential for salvaging ischemic myocardium (Maroko et al., 1971, 1977; Braunwald et al., 1974; Sobel and Shell, 1973; Jugdutt et al., 1979a, 1979b, 1980). Con-
versely, a sharp interface or abrupt gradient of blood flow between or nonoccluded and occluded regions suggests little or no functional anastomoses between intramural branches of the coronary vasculature at the microcirculation level and, consequently, little potential for preferential salvage of the ischemic myocardium at the margins of the ischemic zone (Reimer and Jennings, 1979; Factor et al., 1978, 1981; Hirzel et al., 1977; Yellon et al., 1981).

The present study evaluates the transitional or border zone of intermediate blood flow reduction between nonischemic and ischemic regions following acute coronary artery occlusion in chronically instrumented dogs using methods that minimize an admixture of ischemic and nonischemic myocardium in the tissue analyzed. The regions perfused by occluded and nonoccluded vessels were identified by tracing the extramural and intramural distribution of the coronary vasculature from postmortem angiograms. Regional blood flow was evaluated in serial 3-mm-wide epicardial and endocardial zones from outside and inside the interface between occluded and nonoccluded vessels to provide fine resolution of the distribution of blood flow.

Methods

Studies were performed on 13 mongrel dogs weighing 15-25 kg. The dogs were anesthetized with thialmylal sodium (30-40 mg/kg, iv), and underwent a left thoracotomy. Heparin-filled polyvinyl chloride catheters were inserted into the left atrial cavity and the aortic root and were tunneled to a subcutaneous pouch at the base of the neck. A pneumatic cuff occluder was placed around the left circumflex coronary artery either proximal (10 dogs) or immediately distal (3 dogs) to the first marginal branch and was tunneled to the pouch containing the catheters. Studies were performed 7-10 days after surgery. The pouch was anesthetized with 50 mg lidocaine and the catheters and snare were exteriorized. Phasic and mean aortic and left atrial pressures and an electrocardiogram of lead 2 or 3 were recorded on a Sanborn eight-channel recorder.

Myocardial blood flow was determined by injecting carbonized microspheres 9 ± 1 (so) μm in diameter and labeled with γ-emitting nuclides. The microspheres were obtained as 1 mCi of each nuclide in 10 ml of 10% dextran and 0.05% polysorbate 80 (3M Co.), and were diluted in 10% dextran such that 1.0 ml contained approximately 3.0 × 10⁶ microspheres. Before each injection, the microspheres were mixed by alternate agitation for at least 15 minutes in an ultrasonic bath (3M Co., model DA 0950) and a Vortex agitator. A volume of 1.0 ml of the microsphere suspension was injected into the left atrium over a period of 10-15 seconds, and the atrial catheter was flushed with 5 ml of isotonic saline. Beginning simultaneously with each microsphere injection and continuing for 90 seconds, a reference blood sample was collected in counting vials from the aortic catheter at a constant rate using a Harvard withdrawal pump. Serial injections of the microspheres resulted in no change in heart rate during the interval of injection and no change in aortic or left atrial pressure measured immediately before and after collection of the reference blood samples. After a control blood flow measurement, the left circumflex coronary artery was completely occluded by inflating the pneumatic cuff occluder. One minute later, myocardial blood flow was measured. All dogs were then anesthetized with thialmylal sodium and the hearts were fibrillated with concentrated potassium chloride. The hearts were excised immediately, and the left main coronary artery was cannulated and perfused with barium sulfate gel (Fulton, 1963; Schaper, 1971). The occluder was released during the perfusion. The hearts then were placed in 10% buffered formalin for 3 days to allow fixation.

The epicardial distribution of the coronary vascular branches was determined from gross examination and from radiographs of the entire left ventricle. Each left ventricle was then cut into eight rings from base to apex, as illustrated in Figure 1, and radiographs were made of each section. Figure 1A is a schematic of a radiograph of one ring at the level of the papillary muscle. From radiographs of each transverse section, the intramural distribution of each vessel was traced through the myocardium in a fashion comparable to the branches of a tree. Transmural lines were drawn on the radiographs equidistant between medial and lateral terminal branches of the occluded and nonoccluded vessels. The angulation of the cuts varied to best conform to the distribution of the intramural vessels. These lines were used as a guide for transmural cuts that separated the occluded from the nonoccluded region, as illustrated in Figure 1B. The weights of occluded and nonoccluded regions were determined. The rings were then cut into serial 3-mm-wide sections as illustrated in Figure 1B. Each 3-mm-wide section was cut into epicardial and endocardial halves. Sections from each ring were combined so that myocardial blood flow could be determined in serial 3-mm zones, 1-3 g in size, inside and outside the occluded region. The base ring 1 and apex rings 7 and 8 were not included in the multiple sections and combined analyses. Ring 1 contains fibrous tissue from the valve rings. The apex rings are supplied primarily by the left anterior descending coronary artery. The myocardium supplied by the circumflex artery tapers at the apex to supply a relatively small and variable amount of rings 7 and 8; separation of occluded and nonoccluded vasculature was more difficult in the apex rings. The present

![Figure 1](http://circres.ahajournals.org/)

**Figure 1.** Schematic of the left ventricle with the right ventricle removed. Part A is a schematic of a radiograph of the vasculature injected with barium sulfate gel from one of the eight transverse rings at the level of the papillary muscle. Part B is a schematic of the technique for sectioning each transverse ring. C = nonischemic region, IC = ischemic zone center, Sep = septum, Cir = circumflex, LAD = left anterior descending.
study assesses blood flow gradients at the lateral and medial margins of the occluded vasculature which represents the majority of the interface between the ischemic and nonischemic regions.

The number of zones from the occluded region ranged from 2 to 5 at each border, depending on the size of the region. Two 3-mm zones were cut from the nonoccluded region at each border. Samples were taken from the center of the occluded and nonoccluded regions. The radioactivity in each reference blood and tissue sample was measured in a Packard Gamma Scintillation Spectrometer, using window settings selected to correspond with the peak energies of each radioactive nuclide. Blood flow per tissue sample was determined using the counts per ml per minute for the blood samples and the counts per minute for the tissue sample. Blood flow was calculated using the following formula:

\[ Q_m = Q_r \cdot \frac{C_m}{C_r} \]

where \( Q_m \) = myocardial blood flow (ml/min), \( Q_r \) = reference blood flow (ml/min), \( C_m \) = counts/min in myocardium, and \( C_r \) = counts/min in reference blood flow. Myocardial blood flow (ml/min) was divided by the weight of the zone and was expressed as ml/min per g.

Blood flow measurements in serial zones outside and inside the occluded regions were analyzed in two ways. Blood flow in each 3-mm epicardial and endocardial zone was expressed as a fraction of anterior region flow in each dog; the mean distribution of blood flow for the group then was analyzed. To allow comparison of the transitional zone of intermediate blood flow reduction between individual animals, the maximum blood flow gradient between nonischemic and ischemic tissue was determined in each animal by subtracting the lowest blood flow value in the ischemic zone from blood flow in the anterior nonischemic zone. Myocardial blood flow in each 3-mm zone outside and inside the ischemic region was then expressed as a percent of the maximum blood flow gradient between nonischemic and maximally ischemic regions. All data points from the 13 dogs are illustrated in Figures 3 and 4.

Additional studies were performed on four mongrel dogs to determine to what extent blood flow measurements to the occluded circumflex region resulted from an admixture of normal myocardium. This analysis was accomplished by perfusing the circumflex coronary artery using a modification of the technique of Hirzel et al. (1977) that allowed exclusion of microspheres injected into the left atrium from the vascular distribution of the circumflex coronary artery while perfusion was maintained. Each dog was anesthetized with thiotal sodium (30 to 40 mg/kg, iv) and ventilated with 100% oxygen. A left thoracotomy was performed. The pericardium was incised and the heart suspended in a pericardial cradle. Catheters were filled with heparinized saline and placed in the aortic root and left atrial appendage. The proximal left circumflex coronary artery was isolated proximal to the first marginal branch. Myocardial blood flow was measured during total occlusion as previously described; the occlusion was then released. A second inlet to the reservoir was connected to a balloon within the reservoir so that femoral artery blood could be delivered selectively into either the reservoir or the balloon without changing pressure in the circumflex catheter. After a 15-minute steady state period, femoral arterial inflow was switched to the balloon and microspheres were injected into the left atrium as previously described for analysis of regional blood flow. This arrangement allowed perfusion of the circumflex artery at aortic perfusion pressures from blood in the reservoir and collection of microspheres in the balloon. Immediately after the blood flow measurement, the heart was fibrillated with potassium chloride, excised, perfused with barium sulfate gel, and placed in 10% buffered formalin for fixation, and subsequently sectioned according to the risk region as previously described. The distribution of microspheres through the latter procedure represents the distribution of the vasculature exclusive of the circumflex artery; microspheres present in samples taken from the circumflex region represent the extent of admixture or overlap of nonischemic myocardium in the region.

Results

Figure 2 is a graph of blood flow in ml/min per g in serial 3-mm sections immediately inside and outside the region supplied by the occluded circumflex vessel in a representative study. The region of intermediate flow reduction occurred in a 3-mm-wide section immediately inside the occluded region at both the medial and lateral margins of the ischemic zone. In the nonoccluded region, endocardial flow was greater than epicardial flow; in the occluded zone, epicardial flow was greater than endocardial flow. In the total group, blood flow was reduced to 61 ± 5% and 58 ± 6%, of nonischemic region flow at the medial and lateral epicardial margins, respectively, and 45 ± 6% and 47 ± 5% at the medial and lateral endocardial margins, respectively. Blood flow to the second 3-mm zone inside the occluded region and the central ischemic region were not significantly different.

\[ Q_m = Q_r \cdot \frac{C_m}{C_r} \]

where \( Q_m \) = myocardial blood flow (ml/min), \( Q_r \) = reference blood flow (ml/min), \( C_m \) = counts/min in myocardium, and \( C_r \) = counts/min in reference blood flow. Myocardial blood flow (ml/min) was divided by the weight of the zone and was expressed as ml/min per g.

Blood flow measurements in serial zones outside and inside the occluded regions were analyzed in two ways. Blood flow in each 3-mm epicardial and endocardial zone was expressed as a fraction of anterior region flow in each dog; the mean distribution of blood flow for the group then was analyzed. To allow comparison of the transitional zone of intermediate blood flow reduction between individual animals, the maximum blood flow gradient between nonischemic and ischemic tissue was determined in each animal by subtracting the lowest blood flow value in the ischemic zone from blood flow in the anterior nonischemic zone. Myocardial blood flow in each 3-mm zone outside and inside the ischemic region was then expressed as a percent of the maximum blood flow gradient between nonischemic and maximally ischemic regions. All data points from the 13 dogs are illustrated in Figures 3 and 4.

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Results

Figure 2 is a graph of blood flow in ml/min per g in serial 3-mm sections immediately inside and outside the region supplied by the occluded circumflex vessel in a representative study. The region of intermediate flow reduction occurred in a 3-mm-wide section immediately inside the occluded region at both the medial and lateral margins of the ischemic zone. In the nonoccluded region, endocardial flow was greater than epicardial flow; in the occluded zone, epicardial flow was greater than endocardial flow. In the total group, blood flow was reduced to 61 ± 5% and 58 ± 6%, of nonischemic region flow at the medial and lateral epicardial margins, respectively, and 45 ± 6% and 47 ± 5% at the medial and lateral endocardial margins, respectively. Blood flow to the second 3-mm zone inside the occluded region and the central ischemic region were not significantly different.

\[ Q_m = Q_r \cdot \frac{C_m}{C_r} \]
FIGURE 3. Individual and mean epicardial blood flow measurements from 13 dogs expressed as a percent of the gradient between nonischemic and maximally ischemic regions. Vertical lines indicate the section between nonoccluded and occluded regions. IC = ischemic zone center.

Transmural gradients in the nonischemic and ischemic regions also were similar to those illustrated in Figure 2.

FIGURE 4. Individual and mean endocardial blood flow measurements from 13 dogs expressed as a percent of the gradient between nonischemic and maximally ischemic regions. The vertical lines indicate the section between nonoccluded and occluded regions. IC = ischemic zone center.

Blood flow reduction in the first 3-mm-wide zone at the medial and lateral epicardial margins of the ischemic region averaged 46 ± 19% and 45 ± 24%, respectively, of the maximum gradient between nonischemic and maximally ischemic regions. There was considerable variation in the blood flow reductions in these regions; the reduction ranged from 11 to 100%. The blood flow reductions in the second 3-mm-wide zone at the medial and lateral epicardial margins of the ischemic region averaged 82 ± 13% and 82 ± 16%, respectively, of the gradient between nonischemic and maximally ischemic zones. The variation in this region was less; the blood flow reduction was less than 70% of the gradient between nonischemic and maximally ischemic zones in only 4 of 26 instances.

Blood flow reduction in the first 3-mm-wide zone at the medial and lateral endocardial margins of the ischemic region averaged 58 ± 23% and 54 ± 17%, respectively, of the gradient between nonischemic and maximally ischemic zones in 11 of 26 instances, the reduction exceeded 60% of the maximum blood flow gradient in the first 3-mm-wide zone. The blood flow reduction in the second 3-mm zone was less than 80% of the gradient between the nonischemic and maximally ischemic zones in only 2 of 26 instances. Blood flow to epicardial and endocardial zones immediately outside the ischemic region was not significantly different from anterior region blood flow. Thus, following acute coronary artery occlusion, the transitional zone of intermediate reduction in blood flow occurred almost exclusively in the first 3-mm zone immediately inside the ischemic region in both the epicardial and endocardial layers.

Table 1 contains blood flow data in ml/min per g in epicardial and endocardial layers from the nonischemic anterior wall and from the center of the ischemic zone. Blood flow to the central ischemic zone is also listed as a percentage of anterior nonischemic blood flow. Blood flow to the epicardial layers of the central ischemic zone was highly variable between animals; mean flow was 0.15 ml/min per g, range 0.07–0.32 ml/min per g, representing an average reduction to 17%, range 7–37% of nonischemic anterior flow. Blood flow to the endocardial layers of the central ischemic zone was reduced to a greater extent; mean flow was 0.06 ml/min per g, range 0–0.16 ml/min per g representing an average reduction to 6%, range 0–17% of the nonischemic blood flow. These data demonstrate significant transmural differences in blood flow between the epicardial and endocardial layers in individual animals and considerable variability between animals in the transmural distribution of blood flow. Thus, although gradients of blood flow at the margins of the ischemic zone occurred over a narrow zone of 3 mm or less, substantial differences in the transmural distribution of blood flow were present between the epicardial and endocardial layers of the remaining ischemic zone.
The total ischemic or risk region in the present study averaged 38.8% of the left ventricular weight, range 25.5–54.6%. The 3-mm-wide zones of intermediate reduction in blood flow at the border of the ischemic zone averaged 12.9%, range 7–19% of rings 2–6.

Figures 5 and 6 illustrate the distribution of blood flow in four dogs expressed as a percent of normal zone flow, between occluded and nonoccluded regions during first acute ischemia and then, during perfusion of the previously occluded vessel, via a two-chamber reservoir bottle that allowed exclusion of microspheres from the perfusate. Blood flow values in the occluded region during no ischemia are a result of an overlap or admixture of normal myocardium in the sample. The difference between blood flow measurements in the occluded region during ischemia and no ischemia is a measure of collateral blood flow at this interval. Table 2 lists blood flow expressed as a percent of normal zone flow in the 3-mm zone immediately inside the occluded region during the two blood flow measurements; the percent of flow during ischemia that may be explained by overlap or an admixture of nonischemic myocardium is tabulated. During no ischemia, blood flow averaged 37% and 30% of normal zone flow in the medial and lateral epicardial regions, respectively, and 33% and 36% in the medial and lateral endocardial regions, respectively, in the first 3-mm region inside the occluded region. The overlap of nonischemic myocardium in the occluded region was limited primarily to the first 3-mm section; overlap blood flow averaged 6 and 3% in the epicardial and endocardial regions of the second 3-mm section; no overlap blood flow occurred in the remaining sections from the occluded region. These data indicate that the intermediate zone of reduced blood flow in these studies resulted primarily from an admixture of nonischemic myocardium.

### Table 1

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>Mean ± SD</th>
<th>Nonischemic (ml/min per g)</th>
<th>Nonischemic blood flow (% nonischemic)</th>
<th>Central ischemic zone (ml/min per g)</th>
<th>Central ischemic zone (% nonischemic)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td>0.89 ± 0.31</td>
<td>10</td>
<td>0.99</td>
<td>4</td>
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<tr>
<td>2</td>
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<td>0.78 ± 0.13</td>
<td>17</td>
<td>0.86</td>
<td>15</td>
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<tr>
<td>3</td>
<td></td>
<td>1.02 ± 0.20</td>
<td>20</td>
<td>1.14</td>
<td>8</td>
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<tr>
<td>4</td>
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<td>0.91 ± 0.18</td>
<td>20</td>
<td>1.01</td>
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<tr>
<td>5</td>
<td></td>
<td>1.22 ± 0.11</td>
<td>9</td>
<td>1.32</td>
<td>2</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td>0.48 ± 0.12</td>
<td>25</td>
<td>0.64</td>
<td>8</td>
</tr>
<tr>
<td>7</td>
<td></td>
<td>0.74 ± 0.07</td>
<td>9</td>
<td>0.92</td>
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<tr>
<td>8</td>
<td></td>
<td>0.87 ± 0.32</td>
<td>37</td>
<td>0.95</td>
<td>17</td>
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<td>9</td>
<td></td>
<td>0.60 ± 0.17</td>
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Mean ± SD: 0.95 ± 0.31, 0.15 ± 0.07, 17 ± 9, 1.06 ± 0.27, 0.06 ± 0.05, 6 ± 6

### Discussion

The present study analyzes the distribution of blood flow between nonoccluded and occluded myocardial regions. Postmortem angiograms were used as a guide to identifying the epicardial and transmural distribution of the nonoccluded and occluded vasculature. Serial 3-mm sections were taken from inside and outside the distribution of the occluded vasculature to provide fine resolution of the distribution of blood flow. The results of these analyses demonstrate that intermediate reductions in blood flow between nonischemic and central ischemic regions occurred in a very narrow rim at the outer margin of the occluded region. An additional group of studies was carried out to determine to what extent the zone of intermediate blood flow reduction represented an admixture of nonischemic and ischemic myocardium. These studies utilized a two-chamber reservoir that allowed exclusion of microspheres from the distribution of the circumflex vasculature while maintaining perfusion as described by Hirzel et al. (1977). The results of the latter study indicate that most of the intermediate reduction in blood flow resulted from an admixture of nonischemic myocardium in the sample. These studies support the view that the blood flow gradient between nonischemic and acutely ischemic myocardium is abrupt, with essentially no significant zone of intermediate ischemia. Overlap of nonischemic myocardium in the circumflex or occluded region was limited primarily to the first 3-mm section; overlap in the second 3-mm section averaged 6% and 3% in the epicardial and endocardial regions; there was no overlap in the remaining sections of the occluded zone. Thus, the dimensions of the zone of admixture or interdigitation of normal and ischemic myocardium was relatively small in the sections analyzed. These data indicate that the postmortem angiographic technique provides...
a guide to identify the distribution of the nonoccluded and occluded vasculature and thus to delineation of the ischemic zone. The technique minimizes the degree of overlap of ischemic and nonischemic tissue to a narrow border at the margins of the ischemic zone. Because of the interdigitation of nonischemic and ischemic myocardium in this narrow zone, the technique does not completely separate myocardium perfused by the two circulations in this narrow segment. Admixture of normal and ischemic myocardium may result from interdigitation in a given plane as well as from overlap at different depths through the section. The x-ray angiogram should thus be made from myocardial sections that are relatively thin. In the present study, the left ventricle was sectioned into eight rings; this resulted in small epi-end samples that were then combined from different rings to provide a sample size greater than 1 g.

The view that a zone of intermediate ischemia and injury constitutes a significant fraction of the ischemic region is supported by a variety of studies [histochemical (Vokonas et al., 1978; Cox et al., 1968), biochemical (Cox et al., 1968; Hearse et al., 1977; Kjekshus and Sobel, 1970), myocardial blood flow (Becker et al., 1973; Vokonas et al., 1978; Jugdutt et al., 1979a, 1979b, 1980), and electrocardiographic (Maroko et al., 1971, 1977; Braunwald et al., 1974)]. It is apparent that intermediate reductions in blood flow or injury may result if a tissue sample that is analyzed contains an admixture of nonischemic and ischemic myocardium and that the dimension of the zone will depend on the technique for sampling. Most of the aforementioned studies did not section the ventricle in a fashion that would allow fine resolution of the dimension of the interface between occluded and nonoccluded regions and did not consider the extent of admixture of nonischemic and ischemic tissue in samples analyzed.

Certain studies that have analyzed the distribution of infarction as a function of the occluded vasculature identified by different dye injections (Reimer et al., 1979) or postmortem angiograms (Koyanagi et al., 1982) have observed that infarction at the endocardial surface extended to 1-2 mm or less of the lateral margins of the occluded vasculature at 4 days to 24 hours after coronary occlusion; although these studies support sharp separation between the occluded and nonoccluded vasculature, especially in the endocardium, the distribution of blood flow was not analyzed at the interface between occluded and nonoccluded regions following acute occlusion. Studies by Kirk and associates (Factor et al., 1978, 1981; Hirzel et al., 1977) have supported the concept of a sharp or abrupt,
rather than gradual, transition between nonischemic and ischemic tissue 24 hours after coronary occlusion. Hirzel et al. (1977) observed no lateral border zone of intermediate creatine kinase depletion 24 hours after coronary occlusion in dogs when tissue supplied by the nonoccluded vasculature was excluded from the analyses. In subsequent studies from the same laboratory, Factor et al. (1978) reconstructed the three-dimensional geometry of the border area from serial sections 24 hours after coronary occlusion in the dog. They observed an irregular interface between normal and infarcted myocardium; apparent islands of normal tissue in the ischemic zone resulted from interdigitating peninsulas of normal tissue. Okun et al.

### Table 2

<table>
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<th>Dog no.</th>
<th>Ischemia</th>
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<th>% Overlap</th>
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<td>Mean ± sd</td>
<td>46 ± 13</td>
<td>30 ± 11</td>
<td>65 ± 12</td>
<td>40 ± 19</td>
<td>36 ± 20</td>
<td>86 ± 9</td>
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FIGURE 6. Blood flow in 3-mm epicardial and endocardial zones from inside and outside circumflex regions during circumflex ischemia (open circles) and no ischemia (closed circles) in dogs 3 and 4. Blood flow is expressed as a fraction of anterior region flow. Microspheres were excluded from the circumflex vasculature during no ischemia. Vertical lines indicate the sections between nonoccluded and occluded regions. IV = ischemic zone center.
During the occlusion period, the blood flow to the ischemic myocardium is measured to assess the extent of injury. Studies using microangiography in acute and chronic settings have shown that the ischemic zone is surrounded by a narrow rim of myocardium that is neither entirely ischemic nor fully perfused.

Analysis of transmural blood flow within the ischemic zone demonstrated significant differences in blood flow between the endocardial and epicardial layers in individual animals and considerable variation in a given layer among animals. Epicardial flow to the center of the ischemic region varied from 7 to 37%, average 17% of blood flow to anterior nonischemic regions. Endocardial blood flow to the center of the ischemic region varied from 0 to 17%, average 6% of nonischemic blood flow. Since the studies performed in acutely anesthetized dogs indicate that admixture of nonischemic myocardium did not extend past the second 3-mm zone inside the occluded region, these variations in blood flow result from differences in collateral blood flow and thus represent differences in the degree of ischemia.

As noted in the previous discussion, numerous methods have been utilized to characterize the transitional or border zone of blood flow and/or ischemic injury after coronary occlusion in acute open-chest studies. Potential advantages of the angiographic technique (Fulton, 1965; Schaper, 1971) are that it is a postmortem method that does not require instrumentation of the intact vasculature and thus is applicable for use in studies that are performed in chronically instrumented as well as acute prepartations; it should not be influenced by the development of epicardial collateral vessels following coronary artery occlusion and infarction, and it provides a logical anatomic basis for sectioning the ventricle for analysis of blood flow. As discussed previously, the technique minimizes overlap of normal and ischemic tissue to a narrow zone at the outer rim of the occluded region, but does not allow complete separation of nonischemic and ischemic tissue. The injection of different colored dyes to stain myocardial regions supplied by occluded and nonoccluded vessels is another technique that has been used to identify the ischemic zone (Reimer and Jennings, 1979). There are several potential sources of error that may be encountered in using dye injection technique. The dye technique requires simultaneous perfusion of the occluded and nonoccluded vessels at equal pressures to minimize passage of dye via the collateral vessels. Differential pressures may result from simultaneous perfusion of different sized vascular beds, kinked vessels at the catheter sites, different sized perfusion catheters, and/or underperfusion of one or more of the major branches of the left main coronary artery that often arise from immediate trifurcations into septal, anterior descending, and circumflex branches. These potential problems are greater in the presence of enhanced collateral vessel development. Permanent occlusion causes rapid development of epicardial collateral vessels (Gregg, 1974) so that 3 days after coronary occlusion, dye, as well as radiopaque barium gel, injected into nonoccluded vessels may easily pass through the epicardial collaterals and stain or fill the ischemic as well as nonischemic myocardium and vasculature. The barium gel technique used in the present study depends on filling of the entire vasculature; simultaneous perfusion of occluded and nonoccluded vessels is not required, and the presence of collaterals facilitates filling, should a branch be underperfused.

In summary, an analysis of blood flow between ischemic and nonischemic regions, based on the distribution of the intramural coronary vasculature from postmortem angiograms, demonstrated that intermediate reductions in blood flow occurred in a very narrow rim of myocardium 3 mm or less in dimension immediately inside the ischemic zone; significant differences in blood flow in the epicardium and the endocardium were present in the remaining ischemic zone. Studies performed in anesthetized dogs in...
which microspheres were excluded from the circumflex regions during no ischemia indicate that the intermediate reductions in blood flow at the border of the ischemic region result primarily from an admixture of normal myocardium and thus do not represent a border zone of intermediate ischemia.

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INDEX TERMS: Ischemic border zone • Transitional blood flow zones • Acute myocardial ischemia • Admixture of myocardial tissue
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