Myocardial oxygen consumption, oxygen supply/demand heterogeneity, and microvascular patency in regionally stunned myocardium

LLOYD D. STAHL, M.D.,* HARVEY R. WEISS, PH.D., AND LEWIS C. BECKER, M.D.

ABSTRACT Although oxygen consumption closely parallels mechanical work in the normal heart, previous studies have found that stunned myocardium may have normal or even increased oxygen consumption despite depressed function. In this study we used microspectrophotometry to measure the oxygen saturations within arteries and veins of less than 100 μm diameter in quick-frozen biopsy samples from normal and regionally stunned myocardium of 10 open-chest anesthetized dogs. Regional myocardial blood flow, measured by radioactive microspheres, was similar in stunned and normal regions, as was mean arteriolar oxygen saturation. However, mean venous oxygen saturation was lower in the stunned region (epicardium 38.0% vs 43.8%, p < .02; endocardium 36.2% vs 39.5%, p = .12), indicating increased oxygen extraction and consumption, despite a marked reduction in mean systolic segmental shortening from 14.4% to 0.5%. In addition, there was greater vein-to-vein heterogeneity of oxygen saturation in the stunned region, with an excess of veins having low saturations (statistically significant in epicardium, nonsignificant trend in endocardium). Microvascular injection studies with Microfil or drafting ink revealed filling of over 95% of arterioles and 85% of capillaries in the stunned region, similar to the findings in the normal region. Our results are consistent with an inefficient transfer of energy into myocyte contraction or an increased use of energy for noncontractile activities in stunned myocardium. In addition, the finding of increased heterogeneity of oxygen extraction suggests that the injury to stunned myocardium may not be uniform to all contractile elements, but instead may be focally and irregularly distributed.


REPERFUSION after a brief episode of myocardial ischemia may result in prolonged dysfunction without necrosis, a phenomenon now known as "myocardial stunning."1-3 Since myocardial oxygen consumption closely parallels energy production and mechanical work in the normal heart,4-6 one would anticipate that stunned myocardium should exhibit reduced oxygen consumption. Although several abstracts have appeared describing reduced7,8 normal,9,10 or even increased11 oxygen consumption in stunned myocardium, the only published study bearing on this issue indicates that postischemic myocardial oxygen utilization may be increased above normal.12 Following reperfusion after 2 hr of hypothermic cardioplegic ischemia, canine hearts demonstrated normal pressure-volume curves, but a 40% increase in oxygen utilization at matched levels of developed pressure.12

Although the cause of myocardial stunning is unknown, it has been suggested that calcium overload may represent a major factor.13 Intracellular sodium, which accumulates during ischemia, may rapidly exchange for calcium during reperfusion, as acidosis (an inhibitor of sodium-calcium exchange) resolves. Hoeter et al.14 have shown that increased myocellular calcium content, induced by sodium-calcium exchange via manipulations of perfusate composition, can result in reduced systolic function, while myocardial oxygen consumption is paradoxically increased. The recent finding by Kusuoka et al.13 that reperfusion with low calcium–containing perfusate eliminates myocardial stunning provides further support for the calcium overload hypothesis. Inappropriately high oxygen consumption, if present, could reflect an increased utili-
zation of energy for movement of calcium in stunned myocardium.

Virtually nothing is known about the homogeneity of abnormalities within stunned myocardium. It is unclear whether the dysfunction of a stunned myocardial segment represents uniform depression of all contractile elements, or patchy, focally distributed injury. Information on this issue is critical for an understanding of the pathophysiology and pathogenesis of stunned myocardium.

In this study we used the technique of microspectrophotometry to compare oxygen extraction and consumption in stunned and normal myocardium by determining the oxygen saturation of blood in small arteries and veins less than 100 μm in diameter in quick-frozen myocardial biopsy samples taken from regionally stunned dog hearts. The microscopic nature of this technique allowed us to measure the vein-to-vein uniformity of oxygen extraction within the stunned and normal myocardial segments. In addition, we assessed the patency of the microvasculature in stunned and normal myocardium by microscopic analysis of hearts infused in situ with Microfil or drafting ink.

Methods

Animal preparation. Fifteen mongrel dogs of either sex (20 to 25 kg) were anesthetized with intravenous sodium thiobital (12.5 mg/kg) and intramuscular α-chloralose (100 mg/kg) in urethane, intubated and ventilated with room air at a constant volume with a piston respirator. After a left thoracotomy, the lungs were retracted and the heart was exposed through a pericardiotomy.

Pairs of 5 MHz cylindrical piezoelectric crystals (0.06 inches outside diameter, 0.06 inches in length, 0.015 inches thick; Vernitron, Columbus, OH) were implanted in the myocardium served by the anterior descending coronary artery and the anterolateral basal region within the circumflex artery territory. The crystals were implanted in the midwall of the left ventricle, 10 to 15 mm apart, and were oriented parallel to the minor axis.

Segmental lengths were measured with a pulse transit sonomicrometer (model Sono-1-XB, James Davis Consultants). Left ventricular pressure, and its first derivative with respect to time, left ventricular dP/dt, were measured with a catheter-tipped pressure transducer (Millar) that was inserted through an implanted silicone rubber left atrial catheter and advanced across the mitral valve. Aortic pressure was measured by inserting a catheter-tipped pressure transducer (Millar) into the aortic root from the right carotid artery. Catheters were placed in the descending aorta for withdrawal of microsphere reference samples, the left atrium for microsphere injections, and the femoral vein for infusion of fluids. Aortic and left ventricular pressures and regional segmental lengths were recorded continuously on a direct-writing recorder (Gould Brush 200).

Experimental protocol. A pneumatic occluder was placed around the left anterior descending coronary artery just distal to the first diagonal branch. Stunning was produced by ten 5 min occlusions alternating with 10 min reflow periods, followed by a 90 min recovery period.

Blood flow to stunned and normal myocardium was determined by radioactive microspheres (15 μm diameter, New England Nuclear Co.) labeled with 141 Ce or 115 Sn. Three to five million microspheres were injected into the left atrium after a 3 min mechanical agitation, while reference arterial samples were withdrawn at a constant rate of 2.16 ml/min by a calibrated pump. Blood flow was calculated from the radioactivity in the tissue and reference blood samples determined in a well-type gamma scintillation counter (Packard model 5986) by standard methods after correction for overlap of the energy peaks. In each dog, flows to stunned and normal zones represented measurements in single tissue blocks weighing 1 to 3 g, divided into endocardial and epicardial halves. Flows were measured at baseline and 90 min after the last ischemic episode.

Hemodynamic and ultrasonic crystal measurements were made at end-expiration during the baseline state and 90 min after the final occlusion. In the first group of 10 dogs, after the stunning protocol, 3 to 5 g transmural biopsy samples were taken with use of a scalpel from the area of the stunned and normal regions encompassed by the sonomicrometer crystal pairs and were immersed immediately in liquid nitrogen. The time from initiation of the biopsy to immersion in liquid nitrogen was less than 10 sec in all cases. The samples were coded and stored in liquid nitrogen for future blinded analysis.

The animals were then killed by intra-atrial injection of potassium chloride. The hearts were cut into five to six short-axis slices from apex to base, each of which was incubated at 37°C in triphenyltetrazolium chloride for 20 min and photographed in color. After this the site of each crystal pair was excised, weighed, and counted for radioactivity for measurement of blood flow. The fresh tissue and photographs were inspected to identify unstained areas that represented myocardial necrosis.

A second group of five dogs underwent the identical instrumentation and repetitive coronary occlusion protocol. Ninety minutes after stunning was produced the sonomicrometer crystal pairs were removed and their locations were marked by loosely tied epicardial purse-string sutures. A 1/4 inch Teflon catheter was inserted through the left subclavian artery and advanced into the aortic root just above the aortic valve. The main pulmonary artery was ligated and the right atrium was clamped to a reservoir. After approximately 3 to 5 sec, the ascending aorta was ligated proximal to the great vessels. In three dogs 50 ml of heparinized normal saline followed by 50 ml of Microfil silicone rubber compound (MV-122 Yellow, Canton Biomedical Products, Inc., Boulder, CO) was infused into the aortic root under 120 mm Hg constant pressure. In two other dogs, 8000 units of intravenous heparin was given before ligation of pulmonary artery. After ligation of the ascending aorta, 50 ml of heparinized normal saline was infused into the aortic root followed by 300 ml of black drafting ink (Pelikan, Hannover, West Germany) at a pressure of 120 mm Hg. All hearts continued to beat through the injection period. The hearts were excised below the atrioventricular ring, frozen immediately in liquid nitrogen, and then stored at 70°C for future analysis.

Analysis of function. Signals from the ultrasonic crystals, left ventricular and aortic pressure, and left ventricular dP/dt were routed to a microcomputer (North-Star Horizon) at selected times during the experiment, digitized at 150 Hz by a Tecomar 12-bit analog-to-digital converter, and recorded on floppy disk for later computer analysis. The end-systolic segment length was measured 20 msec before peak negative left ventricular dP/dt15, end-systolic length was measured just before the onset of positive left ventricular dP/dt. Percent systolic segmental shortening was calculated as segmental shortening (end-diastolic minus end-systolic length) divided by end-diastolic length times 100, and represented the average of 5 heartbeats.

Analysis of oxygen extractions. The quick-frozen biopsy samples from the stunned and normal regions of the dogs in the first group were prepared as follows. Thirty micrometer thick
frozen tissue sections were cut on a Slee automated microtome-cryostat set at −35°C in a nitrogen atmosphere. Each section was then transferred to a precooled slide, covered with degassed silicone oil, and rapidly transferred to the microspectrophotometer cold stage flushed with nitrogen gas. Arteries and veins less than 100 μm in diameter were located and absorbances at 560, 523, 506 nm were obtained to give oxygen saturation of the blood contained within the vessels. Only vessels seen in transverse section were studied, so that the light path was only through blood. Between seven and 10 arterioles and seven to 10 venules in the endocardial and epicardial segments of the stunned and normal regions were analyzed for each dog.

Regional oxygen extraction (ml O2/100 ml blood) was calculated as the difference between mean arterioal and venous oxygen saturation, multiplied by the arterial hemoglobin concentration, times the maximal oxygen-combining capacity of 1.36 ml O2/g hemoglobin. Hemoglobin concentration was determined with a Fisher hemophotometer (Fisher Scientific Co., Pittsburgh) from blood samples obtained at the end of the study. The oxygen consumption for each segment of interest was calculated as the product of oxygen extraction and regional blood flow.

Microscopic evaluation of the microvasculature. To identify perfused microvessels (both Microfil and drafting ink), samples were prepared as follows: 2 μm thick sections were cut from the hearts on a Slee automated microtome-cryostat at −35°C. Eight to twenty sections from the endocardial and epicardial segments of both the normal and stunned regions were cut for each animal. Each section was at least 150 to 200 μm from the previous one. As described previously, the slides were photographed through a Zeiss microscope equipped for automated photography. A 40× planapochromat objective was used to photograph the capillary fields and a 10× objective was used to photograph the fields used for observation of arterioles. The slides were then stained for alkaline phosphatase, as described previously. Briefly, they were fixed in a sucrose-formalin buffer for 1 min, washed twice in distilled water, and then placed in an incubation mixture for 30 min at 37°C. The incubation mixture consisted of 3.8 g/liter of Fast blue RR, 0.5 g/liter of α-naphthylphosphate, 3.8 g/liter of sodium metoborate, and 1.7 g/liter of magnesium sulfate. The slides were postfixed and dried.

Various stereologic determinations were made, counting each field twice, once for the total and once for the perfused microvasculature. The system used was a Dapple image analyzing device. We placed the slides that were stained for alkaline phosphatase under a microscope and the image analyzer evaluated the field for total morphometric indexes of microvascularity. The image was obtained from a Panasonic television camera attached to the microscope and digitized with the Dapple image analyser. The number of microvessels per square millimeter was estimated from the number per unit test area. The diameter of the microvessels was determined by measurement of the minimum diameter of any vessel cut in transverse sections such that the maximum diameter was no more than 1.5 times the minimum. After the total microvasculature was studied, the perfused portion was obtained by analysis of photographs of the field for the presence of either microfil or the carbon particles of the drafting ink.

Data analysis. Data are presented as the means ± SD. Comparisons between the means were by paired t test. Comparisons of proportions in the stunned and normal regions were performed by chi-square analysis.

Results

Hemodynamic changes after repeated brief coronary occlusions. Comparing baseline and 90 min after repeated brief coronary occlusions, there were no signifcant differences in heart rate (128 ± 20 vs 133 ± 26 beats/min), peak left ventricular pressure (124 ± 18 vs 131 ± 13 mm Hg), or mean aortic pressure (100 ± 13 vs 105 ± 10 mm Hg).

The effects of repeated coronary occlusions on regional function. Systolic segmental shortening in the left anterior descending region decreased from 14.4 ± 3.3% at baseline to 0.4 ± ±.6% after the repeated 5 min coronary occlusions (p < .001; table 1). The end-diastolic segmental length increased from 12.2 ± 2.1 mm at baseline to 14.6 ± 2.9 mm after stunning (p < .05). The control region showed no change in systolic segmental shortening (11.0 ± 2.2% at baseline vs 10.7 ± 1.7% after repeated occlusions) or end-diastolic segment length (11.7 ± 1.9 vs 11.5 ± 2.1 mm).

Myocardial necrosis. No areas of necrosis were observed on visual inspection of the triphenyl tetrazolium chloride–stained left ventricular slices from any of the 15 animals.

Regional arterial and venous oxygen saturations and oxygen extraction. The mean arterial and venous oxygen saturations in the stunned and control regions are shown in table 2. One hundred fifty-eight arterioles were sampled from the control regions of the 10 dogs (79 in the endocardial region and 79 in the epicardial region). One hundred sixty-one arterioles were sampled from the biopsy specimens from the stunned region (79 in the endocardial region, 82 in the epicardial region). Values for mean arterial oxygen saturation were somewhat low, ranging from 83.4% to 85.7%, probably reflecting the fact that the dogs were ventilated with room air in the right lateral decubitus position. However, there were no significant differences in the mean arterial oxygen saturation or in the frequency distribution of the arterial saturations in the stunned and control regions.

With respect to venous saturations, a significantly lower mean epicardial venous oxygen saturation was

<table>
<thead>
<tr>
<th>TABLE 1</th>
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<tr>
<td>Regional function and blood flow in stunned and control regions</td>
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<tr>
<td>Stunned region</td>
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<td>Baseline</td>
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<tr>
<td>90 min after ischemia</td>
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<tr>
<td>Control region</td>
</tr>
<tr>
<td>Baseline</td>
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<tr>
<td>90 min after ischemia</td>
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</table>

* p < .001 compared with baseline value; b p < .05 compared with baseline value.
TABLE 2
Mean oxygen saturations, extraction, and consumption in stunned and control regions

<table>
<thead>
<tr>
<th></th>
<th>Stunned</th>
<th></th>
<th>Control</th>
<th></th>
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<tbody>
<tr>
<td></td>
<td>Endo</td>
<td>Epi</td>
<td>Endo</td>
<td>Epi</td>
</tr>
<tr>
<td>Arterial saturation (%)</td>
<td>83.9±3.4</td>
<td>85.7±4.0</td>
<td>83.4±2.9</td>
<td>85.4±4.2</td>
</tr>
<tr>
<td>Venous saturation (%)</td>
<td>36.2±14.6*</td>
<td>38.0±13.9#</td>
<td>39.5±15.3</td>
<td>43.8±11.8</td>
</tr>
<tr>
<td>O₂ extraction (ml O₂/100 ml blood)</td>
<td>7.1±1.0*</td>
<td>7.1±0.9#</td>
<td>6.6±1.0</td>
<td>6.3±0.8</td>
</tr>
<tr>
<td>O₂ consumption (ml O₂/min/100 g)</td>
<td>6.9±1.5</td>
<td>8.8±1.5#</td>
<td>6.4±1.9</td>
<td>7.8±1.3</td>
</tr>
</tbody>
</table>

*P = .12 compared with control value.
#P < .05 compared with control value.

observed in the stunned region compared with control (38.0±13.9% vs 43.8±11.8%, p < .02, paired t test). There was also significantly greater vein-to-vein heterogeneity in the epicardium of the stunned region (figure 1). The proportion of veins with low oxygen saturations (<20%) was higher in the stunned than normal epicardium (14.9% vs 4.5%, p < .05).

In the endocardium of the stunned region, the mean venous oxygen saturation was also lower (36.2±14.6% in the stunned region vs 39.5±15.3% in the control region), although the difference did not reach statistical significance. The frequency distribution showed a tendency toward more veins with lower oxygen saturations in the stunned region (figure 2).

Oxygen extraction was higher in the stunned region (7.1±1.0 vs 6.6±1.0 ml O₂/100 ml blood in the endocardium and 7.1±9 vs 6.3±8 ml O₂/100 ml blood in the epicardium), although the difference reached statistical significance only in the epicardium (p < .05; figures 3 and 4, table 2).

Regional myocardial oxygen consumption, calculated from regional oxygen extraction and blood flow, was about 10% higher in the stunned region (statistically significant in the epicardial segment; figures 3 and 4, table 2). The values for mean oxygen consumption (6.4 to 8.8 ml O₂/min/100 g) were somewhat low, based on both the low arterial oxygen saturation and the low arterial hemoglobin concentration (average 11.25 mg/dl), but were within the range previously reported for open-chest dogs.

There was no difference in the diameters of the vessels identified and used to obtain oxygen saturations in the stunned (mean arterial diameter 29.1±21.2 μm with 87% of the vessels having a diameter of less than 60 μm; mean venous diameter 25.2±21.1 μm with 91% less than 60 μm) and the control regions (mean arterial diameter 29.4±24.6 μm with 89% less than 60 μm; mean venous diameter 24.7±19.3 μm with 91% less than 60 μm).

Evaluation of microvascular patency. To determine whether the shift in venular oxygen saturations toward lower values in stunned myocardium was due to focal microvascular obstruction with heterogeneously impaired perfusion, hearts were injected in situ with either Microfil or drafting ink and evaluated microscopically (tables 3 and 4). In three hearts injected with Microfil
to visualize arterioles, similar numbers of vessels were identified in stunned and control myocardium (epicardium, 0.92 vs 1.03 vessels/mm²; endocardium, 1.79 vs 2.10 vessels/mm²). Approximately twice as many vessels were observed in endocardial samples as in epicardial samples in both regions. The percent of vessels patent (i.e., filled with Microfil) was 96% or more and did not differ in stunned and control myocardium. In two hearts injected with drafting ink to visualize capillary filling, similar numbers of vessels were again identified in stunned and control specimens (epicardium, 3950 vs 4123 vessels/mm²; endocardium, 3411 vs 3455 vessels/mm²). Approximately 85% of vessels were patent (i.e., filled with carbon particles), without any difference between stunned and control regions.

**Discussion**

The major findings of this study were that despite reduced systolic function, regionally stunned myocardium exhibited higher mean oxygen extraction and consumption in the presence of normal mean myocardial blood flow. The stunned myocardium also demonstrated a significantly greater vein-to-vein heterogeneity of oxygen saturations, especially in the epicardial segment, with proportionately more venules demonstrating low oxygen saturations, suggesting that the stunned region contained microregions of either increased oxygen extraction or reduced flow. Microscopic examination of normal and stunned regions, however, revealed the microvasculature to be equally patent without evidence of occlusion or plugging.

The microspectrophotometric analysis of arterial and

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**FIGURE 3.** The mean epicardial oxygen extraction and consumption for each dog, comparing results from the stunned and normal regions. The overall mean extraction and consumption was higher in the stunned than in the normal epicardial region.

**FIGURE 4.** The mean endocardial oxygen extraction and consumption for each dog and the overall means for stunned and normal regions.
venous oxygen saturations has been detailed in previous publications and is accurate to approximately ± 3%. The biopsy samples were frozen in liquid nitrogen within 10 sec of removal and stored in a liquid nitrogen container until microspectrophotometric measures were made. It has previously been shown that oxygen saturations in small arteries and veins, even those less than 50 μm in diameter, are not altered if the samples are frozen within 15 to 30 sec, and that the oxygen saturations are stable for weeks at this temperature. All samples were coded and the microspectrophotometric analyses were performed without knowledge of location within the heart.

The accuracy of our determination of regional oxygen consumption depends not only on the arterial and venous saturations, but also on the measurements of regional blood flow. We used mean blood flow values in endocardial and epicardial samples weighing about 1 g each. Based on the number of spheres injected, the estimated measurement error was less than 5%. In addition, the calculation of mean regional oxygen consumption from the individual venular and arteriolar oxygen saturation data involves the implicit assumption that perfusion is homogeneously distributed. If flow were diminished to microregions of high oxygen extraction and replaced by an equal increase in flow to regions of low extraction, mean oxygen extraction would actually be lower than the result obtained by simple averaging of all of the individual extraction values. In previous studies, the validity of the use of averaged microscopic values to obtain mean oxygen extraction has been established both for the heart and gracilis muscle by demonstrating comparability of the microspectrophotometric method and mean extraction obtained from measurements in large arteries and veins. Unfortunately, the distribution of perfusion cannot be examined directly, since methods are not available to measure flow with sufficiently high spatial resolution in the beating heart.

Our finding that oxygen extraction and consumption were higher in stunned myocardium could theoretically be incorrect if the microvascular distribution of perfusion were grossly different in stunned and normal regions. However, the differences involved would have to be extreme. For example, we found a 13% increase in oxygen extraction and consumption in stunned epicardium. To lower this excess oxygen consumption to normal, flow would have to be absent from veins with the lowest saturation (<20%) and replaced by flow to veins with the highest saturation (>50%; figure 1). Furthermore, if flow in normal myocardium were not completely homogeneous and also had a tendency to be distributed more to veins with higher saturations, the maldistribution in stunned myocardium would have to be even greater. Preliminary results from Dean et al., who used regional coronary venous oxygen sampling, support the notion that oxygen consumption in stunned myocardium is at least equal to normal.

There are a number of questions that arise from the results of this study. First, how does one interpret the finding in stunned myocardium of increased oxygen consumption and normal regional blood flow in the face of markedly depressed regional systolic shortening? Given what appears to be decreased mechanical work being performed by the stunned segment, the measurements of regional oxygen consumption are in marked excess of the metabolic demands attributable to muscle contraction. This suggests an impairment of aerobic metabolic efficiency, or a defect in the conversion of aerobically derived metabolic energy into contractile work.

Other investigators have recently reported normal or increased myocardial oxygen consumption in regionally or globally stunned models, consistent with an impairment of aerobic efficiency. Possible explanations for this phenomenon include uncoupling of oxidative phosphorylation, resulting in inefficient mitochondrial ATP production, or an increase in the cellular energy requirement for processes other than contraction. Mitochondrial uncoupling is

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**TABLE 3**

<table>
<thead>
<tr>
<th></th>
<th>Vessels identified</th>
<th>Number of microvessels</th>
<th>Percent patent</th>
<th>Vessel concentration (vessels/mm² tissue)</th>
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<tr>
<td><strong>Control</strong></td>
<td></td>
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<tr>
<td>Epicardium</td>
<td>128</td>
<td>122</td>
<td>95</td>
<td>1.03</td>
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<tr>
<td>Endocardium</td>
<td>282</td>
<td>271</td>
<td>96</td>
<td>2.10</td>
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<tr>
<td><strong>Stunned</strong></td>
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<td></td>
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<td>Endocardium</td>
<td>236</td>
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<td>1.79</td>
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**TABLE 4**

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<th>Total identified</th>
<th>Perfused</th>
<th>% perfused</th>
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<tbody>
<tr>
<td><strong>Control</strong></td>
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<td></td>
<td></td>
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<tr>
<td>Epicardium</td>
<td>4123 ± 423</td>
<td>3533 ± 365</td>
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<tr>
<td>Endocardium</td>
<td>3455 ± 416</td>
<td>2894 ± 372</td>
<td>84</td>
</tr>
<tr>
<td><strong>Stunned</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epicardium</td>
<td>3950 ± 382</td>
<td>3306 ± 310</td>
<td>84</td>
</tr>
<tr>
<td>Endocardium</td>
<td>3411 ± 281</td>
<td>2872 ± 269</td>
<td>84</td>
</tr>
</tbody>
</table>

*Number of vessels/mm² tissue ± SD.
unlikely, since Edoute et al. have shown that mitochondrial oxidative dysfunction occurs only after a relatively long ischemic exposure, well after the appearance of mitochondrial morphologic changes or postischemic mechanical dysfunction. An increase in noncontractile energy requirements seems a more likely possibility, perhaps related to an increase in ion pumping (i.e., extrusion of calcium from the mitochondria in calcium overloaded myocytes) or an increase in the activity of the calcium ATPase to maintain normal sequestration of calcium by the sarcoplasmic reticulum. Krause and Hess have demonstrated that after only 15 min of normothermic global ischemia in the dog, there is significant depression of sarcoplasmic reticulum function, with reduced calcium transport and depressed Ca\(^{++}\)Mg\(^{++}\)-ATPase activity. Importantly, the coupling between calcium transport and ATPase activity is also impaired, consistent with a need for greater utilization of ATP to move calcium into the sarcoplasmic reticulum.

The finding of normal rather than depressed flow in stunned myocardium is of interest, since one might have anticipated that flow would autoregulate downward in concert with an assumed reduction in metabolic demands. However, our finding of increased oxygen extraction in stunned myocardium suggests that the metabolic demands are actually increased rather than decreased, although the energy produced is not translated into contractile work. Viewed in this light, normal levels of flow in stunned myocardium may actually represent an appropriate level of autoregulation.

A second important question concerns the interpretation of our finding of greater heterogeneity of venous oxygen saturations in stunned myocardium, with proportionately more venules having very low oxygen saturations. In principle, this finding should reflect foci of increased oxygen extraction due to either focally impaired microvascular perfusion or microregions of increased oxygen consumption. In either case, our observation suggests that stunned myocardium may not be affected uniformly. Instead, dysfunction of an entire segment may be related to injury of focal microscopic regions. If stunned myocardium is caused by generation of oxygen free radicals upon reperfusion, as has been suggested by the beneficial effects of free radical scavengers, our data suggest that radical formation may be patchy, possibly related to nonuniform generation within the microvasculature. Based on Engler and Covell's finding that leukocyte depletion results in an attenuation of myocardial stunning, a patchy distribution of leukocytes within the microvasculature may be responsible for a heterogeneous pattern of free radical production and myocyte injury.

The vascular injection studies were performed to gain further insight into the finding of a greater proportion of venules with very low oxygen saturations in stunned myocardium. Our results appear to exclude anatomic vascular obstruction at either the arteriolar or capillary levels as an important mechanism. Because the microfil or drafting ink was injected under physiologic pressures in vivo while the heart was still beating, it is unlikely that the injection process per se caused a significant artifactual reopening of previously obstructed vessels. In addition, microvascular patency is supported by our previous finding of normal maximal flow reserve in stunned myocardium. Hori et al. demonstrated that during progressive occlusion of the microvasculature with microspheres, resting flow initially remained normal due to compensatory focal vasodilation, but the maximal vasodilating capacity of the bed fell progressively. Although anatomic obstructions were not identified, our methods were not designed to exclude functional heterogeneity of perfusion. Although the injections of drafting ink were made "semiphysiologically" and carbon particles were seen in nearly all capillaries, the actual numbers of carbon particles present in each vessel were not quantified. In addition, because the ink was injected over several minutes, even vessels with very slow flow should have been filled.

In conclusion, our results provide new data concerning postischemic regionally stunned myocardium in the open-chest anesthetized dog. We found a significant increase in mean oxygen extraction and consumption despite an apparently marked decrease in contractile function. Although mean perfusion of the stunned myocardium was normal and the microvasculature was normally patent, microspectrophotometry revealed increased heterogeneity with an excess of very low venous oxygen saturations, implying either focally impaired perfusion or increased metabolic activity. These results suggest that stunned myocardium is injured in a nonuniform, patchy fashion, although the precise type of injury incurred and the mechanisms responsible remain to be determined.

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References
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