Myocardial Function in Areas of Heterogeneous Perfusion After Coronary Artery Occlusion in Conscious Dogs

DAVID A. COX, A.B., AND STEPHEN F. VATNER, M.D.

SUMMARY Regional myocardial function and blood flow in endocardial layers were correlated in myocardial segments subtending severely ischemic and adjacent, normally perfused myocardium in conscious dogs 1–3 weeks after recovery from coronary artery occlusion. With coronary artery occlusion induced by a hydraulic occluder, endocardial blood flow (measured with radioactive microspheres) and function (determined with an ultrasonic dimension gauge) in homogeneously nonischemic segments increased slightly but not significantly. In homogeneously ischemic segments, blood flow and function decreased (p < 0.01) by 96 ± 1% and 98 ± 4%, respectively. In segments subtending zones of unequal perfusion, endocardial blood flow increased nonsignificantly in the myocardium surrounding the nonischemic crystal, while decreasing by 93 ± 2% (p < 0.01) in myocardium surrounding the ischemic crystal. Surprisingly, these segments behaved like homogeneously ischemic segments, i.e., endocardial shortening decreased by 92 ± 6% (p < 0.01). Thus, the failure to detect shortening despite normal perfusion of the myocardium surrounding one of the transducers suggests a potential problem with interpretation of regional function measurements or an inability of the apparently nonischemic myocardium to contract.

THE MAGNITUDE of the lateral border zone separating areas of ischemic and normally perfused myocardium after acute coronary artery occlusion has been the focus of controversy. Studies have revealed a discrete boundary between normal and infarcted myocardium,1, 2 and what appeared to be islands of normal myocardium within areas of infarction were actually perinuclear, continuous with the main body of normal myocardium.3 Other studies have also suggested that a large, well-defined, moderately ischemic border zone does not separate severely ischemic and normally perfused myocardium.4, 5

In the present investigation we measured regional myocardial shortening between two transducers subtending adjacent zones of intense ischemia and relatively normal perfusion. Our aim in this study was to determine whether the absolute magnitude of myocardial segment shortening at the ischemic border would be intermediate between that observed in ischemic and nonischemic myocardium or whether more severe dysfunction would occur. This question is important not only for measurements of regional segmental length shortening across tissues of disparate levels of perfusion, but also for wall thickness measurements, where perfusion across the myocardial wall is heterogeneous. This question is also relevant to most clinical techniques used to measure regional myocardial wall motion.

Methods

Fourteen mongrel dogs that weighed 25–40 kg were anesthetized with pentobarbital sodium, 30 mg/kg i.v.

Through a thoracotomy in the fifth left intercostal space, miniature pressure gauges (Konigsberg Instruments) were implanted in the left ventricle through a stab wound in the apex. Doppler flow transducers were implanted proximally on the left circumflex coronary artery or the left anterior descending coronary artery (LAD) in seven dogs each, and hydraulic occluders were placed distal to the flow transducers. Heparin-filled Tygon catheters (Norton Co.) were implanted in the left atrium and descending thoracic aorta. Several pairs of miniature ultrasonic transducers were implanted into the endocardial third of the left ventricular free wall 1–2 cm apart. The average depth of implantation of the transducers into the myocardial wall was approximately 25–30% of the wall thickness as measured from the endocardial surface to the epicardial surface and positioned such that the segments were approximately parallel to the myocardial fibers at that depth according to the data of Streeter et al., 6 with appropriate corrections in orientation made for segments closer to the base and for those closer to the apex of the left ventricle. The transducers were implanted in potentially ischemic zones, in nonischemic zones and in myocardium subtending both zones, such that one transducer was in a potentially ischemic zone and the other in a nonischemic zone (fig. 1).

The miniature pressure gauges were calibrated in vitro and in vivo against Statham P23Db strain-gauge manometers (Statham Instruments) connected to the left atrial and aortic catheters. An improved ultrasonic transit-time dimension gauge7–9 was used to measure regional myocardial segment length. This instrument generates a voltage linearly proportional to the transit time of acoustic impulses traveling at the sonic velocity of approximately 1.5 × 10⁶ mm/sec between the 3-MHz piezoelectric crystals, giving a record of instantaneous myocardial segment length. At a constant room temperature, the thermal drift of the instrument is minimal, i.e., less than 0.01 mm in 6 hours. The frequency response is flat to 60 Hz. Any drift in the measuring system, i.e., the instrument electronics, the data tape recorder, and the oscillograph that displayed

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data, was eliminated by periodic calibrations. This involved substitution of pulses of precisely known duration from a crystal-controlled pulse generator with a basic stability of 0.001%. The position of the miniature ultrasonic transducers was confirmed at autopsy and minimal fibrosis extending < 1 mm from the transducer was observed at the site of implantation. If transducers were not in the endocardial third of the left ventricular wall, the data were discarded.

Regional myocardial blood flow was measured by the radioactive microsphere technique. The microspheres were shipped from the manufacturers (3M Co.) in dry form, in multiple vials for each isotope. The solutions were prepared individually as needed; the microspheres were in contact with the solution for less than 7 days. The concentration of microspheres per milliliter of solution was adjusted to account for natural radioactive decay. The microspheres were suspended in a 0.01% Tween 80 solution (10% dextran) and placed in an ultrasonic bath for 60 minutes. Just before injection, dispersion of the spheres was accomplished by agitation with a vortex mixer. Absence of microsphere aggregation was verified by microscopic examination. Several hours before injection of the microspheres, 1.0 ml of the Tween 80-dextran solution (without microspheres) was injected to determine whether the diluent for the microsphere suspension had an adverse effect on cardiac dynamics. One to 3 million 15-µM microspheres labeled with 46Sc, 99mTc, 85Sr or 141Ce were injected through the catheter implanted in the left atrium for two determinations of blood flow, one during control conditions and one during coronary artery occlusion. A reference sample of arterial blood was withdrawn from the catheter implanted in the aorta beginning 10 seconds before microsphere injection and continuing for at least 2 minutes after the injection had been completed. After the dog had been killed with an overdose of pentobarbital sodium, myocardial samples were obtained from the sites at which regional function was measured by sectioning the myocardium into approximately equal epicardial, midwall and endocardial layers. When the ultrasonic transducers were in the endocardial third of the ventricular wall, the myocardial samples were weighed, placed in a multi-channel gamma well counter (Searle Analytic, Inc.) and counted with appropriately selected energy windows for 10 minutes each.

Experiments were conducted 1–2 weeks after operation with the conscious, unsedated dogs lying quietly on their right sides while control measurements were made of left ventricular pressure, rate of change of pressure (dP/dt), heart rate, and segment shortening and velocity of shortening (dSL/dt) in potentially ischemic zone segments, nonischemic zone segments and segments subtending both zones. After control measurements and the first injection of microspheres were completed, the hydraulic occluder was inflated. Cessation of coronary blood flow was confirmed by Doppler flow measurements. Three of 14 dogs appeared uncomfortable after induction of ischemia, and were given morphine sulfate, 0.25 mg/kg i.v. Measurements were recorded continuously, and the second injection of microspheres was made 10 minutes (n = 2) or 1 hour (n = 12) after coronary artery occlusion. Then the dogs were killed to confirm placement of intramyocardial transducers and obtain myocardial samples for regional blood flow determination. Segments were divided into three groups: (1) normal segments, in which the myocardium surrounding both transducers showed either increases in blood flow after coronary artery occlusion or reductions of less than 25% from control; (2) severely ischemic segments, in which the myocardium surrounding both transducers exhibited reductions in blood flow of greater than 85% from control; and (3) heterogeneously perfused segments, in which the myocardium surrounding one crystal (ischemic) exhibited a flow reduction with coronary artery occlusion of at least 85% and the myocardium surrounding the other (nonischemic) crystal showed either an increase of blood flow or a decrease.
of less than 25%. Fourteen segments (eight in the LAD bed and six in the circumflex bed) met the criteria for nonischemic zone, 42 segments (18 in the LAD bed and 24 in the circumflex bed) met the criteria for ischemic zone and 13 segments in eight dogs (spanning the border between the LAD and circumflex beds) met the criteria for heterogeneous perfusion. In addition, in two dogs, segments were measured adjacent to the ischemic zone in which there were homogeneous increases in blood flow.

Data were recorded on a multichannel tape recorder and played back on two multichannel, direct-writing oscillographs (Gould-Brush) at a paper speed of 100 mm/sec. A cardiactachometer, triggered by the pressure pulse signal, provided instantaneous and continuous records of heart rate. Continuous records of dP/dt and dS/dt were derived from the pressure and segment length signals with Philbrick (Teledyne Philbrick) operational amplifiers connected as differentiators, with frequency responses of 700 and 140 Hz, respectively. A triangular wave signal with known slope (rate of change) was substituted for the pressure and segment length signals to calibrate the differentiators directly.

Values are mean ± SEM throughout. Hemodynamic changes from control were analyzed by the paired t test. Differences in myocardial blood flow among all nonischemic, ischemic and heterogeneously perfused segments were analyzed by two-way analysis of variance.11 Changes from control in regional myocardial function (end-diastolic segment length, end-systolic length, shortening and velocity of shortening) were analyzed by two-way analysis of variance. Differences in regional myocardial function among all nonischemic, ischemic and heterogeneously perfused segments were analyzed by two-way analysis of variance. Finally, the regional flow and function data were analyzed such that for each case in which a given dog contributed more than one segment per group, those values were averaged to yield one value per dog per group to minimize the disparity in sample size. When results were analyzed in this fashion, statistical changes were similar to those obtained when analyzing all segments individually.

Results

Hemodynamic Effects of Coronary Artery Occlusion

When assessed 1 hour after the onset of ischemia, coronary artery occlusion increased heart rate by 26 ± 4% from a control of 96 ± 5 beats/min, mean arterial pressure by 14 ± 4% from a control of 98 ± 3 mm Hg, and left ventricular end-diastolic pressure by 90 ± 23% from a control of 6.9 ± 0.6 mm Hg (all p < 0.01). Left ventricular systolic pressure and dP/dt did not change significantly from the control levels of 119 ± 4 mm Hg and 3210 ± 168 mm Hg/sec, respectively.

Regional Myocardial Blood Flow (fig. 2)

With coronary artery occlusion, endocardial blood flow increased in nonischemic zone segments 24 ± 8% from a control of 1.11 ± 0.11 ml/min/g (p < 0.01), while blood flow increased insignificantly in the myocardium surrounding the nonischemic crystal of the disparate pair, i.e., by 15 ± 12% from a control of 1.22 ± 0.09 ml/min/g. These changes, however, were not significantly different. In marked contrast was the 93 ± 2% reduction in blood flow, from 1.24 ± 0.08 ml/min/g in the myocardium surrounding the other crystal of the disparate pair, a change that was significantly different, by definition, from that in nonischemic segments, but not significantly different from the 96 ± 1% reduction in endocardial blood flow in the ischemic zone segments from a control of 1.14 ± 0.05 ml/min/g.

Regional Myocardial Function (Table 1, fig. 3)

The preocclusion control values for regional myocardial function were not significantly different between the three classes of segments studied.

Homogeneous Nonischemic Segments (n = 14)

With coronary artery occlusion, end-diastolic length increased slightly, by 3.2 ± 1.1%, from a control of 14.5 ± 0.8 mm (p < 0.01). Systolic shortening and velocity of shortening did not change significantly from control values of 2.98 ± 0.24 mm and 29 ± 2 mm/sec, respectively.

Homogeneous Ischemic Segments (n = 42)

Coronary artery occlusion increased end-diastolic length by 5.2 ± 0.6% from a control of 14.9 ± 0.7 mm and reduced systolic shortening by 98 ± 4% from

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<td>Homogeneous nonischemic segments</td>
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<td>End-systolic length (mm)</td>
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<tr>
<td>Heterogeneously perfused segments</td>
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<td>End-diastolic length (mm)</td>
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<td>Systolic shortening (mm)</td>
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<td>Velocity of shortening (mm/sec)</td>
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<td>Homogeneous ischemic segments</td>
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<td>End-diastolic length (mm)</td>
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*Significantly different from preocclusion control (p < 0.01).
FIGURE 2. The effects of coronary artery occlusion on regional myocardial blood flow as a percent change from control for homogeneously nonischemic segments, homogeneously ischemic segments, and heterogeneously perfused segments (middle bars). Asterisks indicate significant changes from control; control values are noted at the base of the bars. Myocardial blood flow changes were similar in the homogeneously nonischemic segments and the myocardium surrounding the nonischemic crystal of heterogeneously perfused segments, whereas homogeneously perfused ischemic segments and the myocardium surrounding the ischemic crystal of the heterogeneously perfused segments underwent similar reductions in blood flow.

FIGURE 3. The effects of coronary artery occlusion on endocardial segment shortening are shown as a percent change from control for homogeneous nonischemic segments, heterogeneously perfused segments and homogeneous ischemic segments. Asterisks indicate significant changes from control. The average control values are listed at the base of the bars. Coronary artery occlusion induced an insignificant increase in nonischemic segment shortening while homogeneously perfused ischemic and heterogeneously perfused segment shortening were reduced similarly.

expected, crystal pairs in zones of homogeneously severe ischemia demonstrated marked reductions (98 ± 4%) in shortening after coronary artery occlusion. Surprisingly, those crystal pairs subtending zones of normal perfusion and severe ischemia behaved almost identically to the homogeneously ischemic segments in terms of regional myocardial function—shortening fell by 92 ± 6%. These findings contrast with zones of homogeneous intermediate perfusion, which show moderate reductions in segment shortening.8 The anatomic distribution of the transducers in the nonischemic, heterogeneously perfused, and homogeneously ischemic segments was similar, i.e., approximately one-half of the segments in each group were located in the LAD bed and one-half in the more posterior and basal left circumflex coronary artery bed. Thus, differences in function among the groups should not reflect differences in regional function between regions of the normal left ventricle.12

These data may explain the different relationships between myocardial blood flow and regional myocardial function observed by some investigators. Gallagher et al.13 and Savage et al.14 found linear relationships between transmural blood flow and regional myocardial wall thickening, while Weintraub et al.15 observed a sigmoidal relationship between endocardial shortening and blood flow. In a previous study from our laboratory,9 we found that an exponential function best fit the relationship between regional blood flow and segment length shortening. In that study, heterogeneously perfused segments were excluded. Since the mean blood flow for such segments would reflect only
moderate ischemia, while the present investigation indicates that they would show severe reductions in function, inclusion of these segments could bias the observed relationship between flow and function.

The data demonstrating abnormal mechanical function despite normal perfusion may be interpreted in many ways. They could indicate a problem with the interpretation of measurements from techniques used to assess regional myocardial function, when two transducers lie in zones of disparate levels of ischemia. The area around the nonischemic crystal may shorten normally, but the area around the ischemic transducer may lengthen so that there is a net lengthening or hypokinesis. It is also likely that normally perfused myocardium adjacent to ischemic myocardium does not function properly, either due to biochemical or mechanical considerations. In support of this concept is a physical model of myocardial infarction developed by Bogen et al.,\textsuperscript{16} which predicted that with acute infarction, regional dyskinesis extends beyond the area of ischemia into nonischemic myocardium. Kerber et al.,\textsuperscript{17} using an echocardiographic device in open-chest anesthetized animals, found that wall motion abnormalities occurred in nonischemic myocardium adjacent to ischemic myocardium. Wyatt et al.\textsuperscript{18} found depression of epicardial shortening in normally perfused regions adjacent to ischemic myocardium. In another study conducted in open-chest dogs, Gallagher et al.\textsuperscript{13} found that 75% reductions in regional wall thickening occurred without significant decreases in transmural blood flow, although subendocardial blood flow was reduced markedly. Thus, wall thickening was compromised severely, despite normal or enhanced perfusion of a significant portion of the wall, i.e., epicardial layers.

Finally, in the present investigation, in two segments adjacent to the ischemic zone where the myocardium surrounding both transducers showed increases in perfusion, function was depressed by 43% and 60%, further supporting the concept that myocardial dysfunction extends beyond the ischemic zone.

We conclude that for measurements of endocardial fiber shortening in areas of markedly heterogeneous perfusion, as measured in this study, as well as for wall thickness measurements where endocardial and epicardial blood flow can differ markedly,\textsuperscript{13} the regional function measurement is governed by the blood flow to the more ischemic transducer of the pair, i.e., transducers subtending nonischemic and ischemic myocardium behave like homogeneously ischemic myocardial segments. This could lead to erroneous interpretation of depressed or absent myocardial function in zones of heterogeneous ischemia, which routinely occurs across the myocardial wall and in potential "border" zones. This problem is relevant to most techniques used clinically to assess regional myocardial function.

References
2. Harken AH, Barlow CH, Harden WR, Chance B: Two and three dimensional display of myocardial ischemic "border zone" in dogs. Am J Cardiol 42: 954, 1978
Myocardial function in areas of heterogeneous perfusion after coronary artery occlusion in conscious dogs.
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