Detection of Coronary Artery Stenosis With Power Doppler Imaging

Flordeliza S. Villanueva, MD; Edward W. Gertz, MD; Melissa Csikari, BS; Gregory Pulido, MS; David Fisher, BS; Jiri Sklenar, PhD

Background—Power Doppler is a new imaging method for detecting microbubbles during myocardial contrast echocardiography (MCE) based on the registration of variance resulting from ultrasound-induced nonlinear bubble behavior. We tested the hypothesis that power Doppler imaging can be used to quantify coronary stenoses.

Methods and Results—Three left anterior descending (LAD) coronary stenoses of varying severity were created in each of 9 open-chest dogs. MCE was performed by continuous intravenous infusion of a nitrogen-filled bilayer shell microbubble, PB127, during triggered power Doppler imaging at incremental pulsing intervals. MCE and radiolabeled microsphere measurements were made at baseline and during each stenosis, with and without adenosine stress. Videointensities in the LAD and left circumflex (LCx) beds were plotted against pulsing interval and fit to a previously described exponential function modeling microbubble destruction and replenishment, which was used to derive parameters of bubble velocity ($\beta$) and peak plateau videointensity (A). Contrast defects matching the location of radiolabeled microsphere hypoperfusion were clearly seen, without need for image processing. The product of $\beta$ and A was linearly related to LAD/LCx flow ($r=0.90, P<0.0001$) and inversely related to stenosis gradient ($r=-0.70, P<0.0001$). Endocardial/epicardial flow ratios were visualized and quantifiable.

Conclusions—As with B-mode harmonics, a model of microbubble destruction/replenishment can be applied to power Doppler data as a means to detect a broad range of stenoses. Image clarity and the lack of attenuation or requirement for background subtraction are additional advantages of this imaging approach. Power Doppler MCE imaging holds promise for the detection of coronary artery disease. (Circulation. 2001;103:2624-2630.)

Key Words: echocardiography ■ contrast media ■ coronary disease

Myocardial contrast echocardiography (MCE) is an imaging method that utilizes intravenously injected microbubbles as red blood cell tracers during simultaneously performed ultrasound imaging.1 The optimal application of MCE for perfusion imaging requires that the contrast agent augment blood echoes and that the imaging system be capable of suppressing the clutter represented by non–contrast-bearing tissue, while at the same time detecting microbubble passage.2

Conventional B-mode ultrasound imaging modified to harmonic mode has been a major innovation in microbubble detection3,4 that has established the potential of MCE for the assessment of myocardial perfusion.3-7 This method, however, is limited by the persistent tissue “noise” consequent to the nonlinear backscatter characteristics of myocardium.

Power Doppler is a new imaging approach for detecting microbubbles that may have advantages over B-mode harmonic imaging.8,9 This adaptation of conventional Doppler technology theoretically displays signals that derive uniquely from ultrasound-induced microbubble disruption.10,11 The premise underlying power Doppler MCE is that acoustically stimulated nonlinear microbubble oscillation or microbubble destruction12-16 results in a changing scatter pattern of the incident ultrasound beam that is detected by a Doppler processor as a large change in the returning radiofrequency signal.10 In power Doppler mode, the autocorrelator of the imaging system calculates the amplitude of the changing signal (as opposed to the frequency shift, as in color Doppler) and displays the amplitude of the signal as the image.10 Because of the strong nonlinear scattering properties of microbubbles,16 the returning signal derives predominantly from the microbubbles rather than from myocardial tissue, which is relatively more acoustically quiescent from the standpoint of a Doppler autocorrelator.

Despite the theoretical appeal of power Doppler for MCE, there are sparse data validating quantitative power Doppler imaging against independent measures of myocardial perfusion in vivo. Whether power Doppler can distinguish between non–flow-limiting stenoses of differing severity or whether quantitative models to assess myocardial perfusion can be
applied to power Doppler images, as has been shown with B-mode harmonics, is unclear. Accordingly, this study was performed to test the hypothesis that power Doppler contrast imaging can detect and quantify coronary artery stenoses.

**Methods**

**Animal Preparation**

A canine model of graded coronary artery stenosis was used. The experimental protocol was approved by the Institutional Animal Care and Use Committee of the University of Pittsburgh and complied with the American Heart Association guidelines for use of animals in research. Nine dogs were premedicated with intravenous sodium pentobarbital (10 mg/kg), intubated, and ventilated. A 20-gauge peripheral intravenous catheter was placed in the forearm for administration of ultrasound contrast. Two 7F catheters were placed in the femoral arteries for pressure monitoring, blood gas analyses, and radiolabeled microsphere references. Catheters were placed in the femoral veins for intravenous medications and fluids. General anesthesia was maintained with sodium pentobarbital.

A left lateral thoracotomy was performed, and the heart was suspended in a pericardial cradle. A catheter was placed in the left atrium for radiolabeled microsphere injection. A peripheral balloon occluder (Research Products International Corp) was placed around the proximal left anterior descending artery (LAD), and transist time flow probes (Transonic Inc) were placed on the midportion of the LAD and left circumflex artery (LCx). A 25-gauge catheter was placed in the distal LAD for measurement of poststenotic pressure.

**Ultrasound Contrast Agent**

The ultrasound contrast agent study used for this study, PB127 (POINT Biomedical Corporation), is specifically engineered for power Doppler imaging and consists of nitrogen encapsulated by a spherical bilayer shell composed of human albumin (outer shell) and a biodegradable polymer (inner shell). The mean diameter is 4.4 ± 2.1 μm, and concentration is 0.5 × 10^9 bubbles/mL; for continuous infusion, 4 mL of the agent was diluted in 50 mL of 5% dextrose water.

**Myocardial Contrast Echocardiography**

Open-chest power Doppler imaging was performed with a commercially available scanner (Harmonic Angio Mode, SONOS 5500, Agilent Technologies, Inc). Machine settings included the following: large packet size, medium line density, pulse repetition frequency of 3.7 to 4.2 kHz, and a mechanical index of 0.8 to 1.0. A yellow-scale color map (“S” map) was used, and B-mode harmonic data were excluded from the digital data set so as to generate a pure Doppler image. Mid-papillary-muscle short-axis images were obtained at each stenosis stage. The transstenotic gradient (difference between mean aortic pressure and distal LAD pressure) of 5 to 10 mm Hg. To create a moderate non–flow-limiting stenosis, the occluder was initially inflated until epicardial flow began to decrease to define the maximal non–flow-limiting gradient, and then partially deflated to result in a stenosis with a gradient 5 to 7 mm Hg less than the maximal gradient. To produce a flow-limiting stenosis, the occluder was inflated until a 50% to 60% reduction in epicardial flow occurred. MCE was performed at each stage, with and without adenosine (0.4 mg · kg⁻¹ · min⁻¹). Radiolabeled microsphere measurements were made during adenosine infusion at each stenosis stage and without adenosine during the moderate stenosis stage. The transstenotic gradient was continuously monitored, and MCE was performed only after at least 5 minutes of hemodynamic stability had been documented after each balloon manipulation. At the end of the experiment, the LAD was totally occluded, India ink was injected to delineate the LAD bed, the dog was euthanized, and the MCE heart slice was sectioned for radiolabeled microsphere analysis.

**Results**

**Severity of Experimental Stenoses**

Radiolabeled microsphere flow to the LAD normalized to LCx bed during the 6 stages in which measurements were taken is shown in Table 1. As expected, at preadenosine baseline, LAD and LCx flows were similar to each other (LAD/LCx ratio 1.01 ± 0.13), and both beds demonstrated a similar hyperemic response to adenosine such that the LAD/LCx flow ratio remained the same (1.10 ± 0.32, P = 0.44).

<table>
<thead>
<tr>
<th>Stage</th>
<th>No Adenosine</th>
<th>Adenosine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>1.01 ± 0.13</td>
<td>1.10 ± 0.32</td>
</tr>
<tr>
<td>Mild stenosis</td>
<td>...</td>
<td>0.47 ± 0.16*</td>
</tr>
<tr>
<td>Moderate stenosis</td>
<td>0.85 ± 0.16</td>
<td>0.31 ± 0.12*</td>
</tr>
<tr>
<td>Severe stenosis</td>
<td>...</td>
<td>0.22 ± 0.09*</td>
</tr>
</tbody>
</table>

Data are expressed as mean ± SD. n = 9.

Ellipses indicate microsphere flow not measured.

*P < 0.001 vs baseline/adenosine.

With increasing stenosis, LAD hyperemic flow in response to adenosine was blunted, resulting in a progressive decrease in red cell velocity and the duration of time before the next destructive pulse. At progressively longer intervals between pulses, microbubbles fully replenish the beam, and a peak plateau videointensity, A, is achieved. Using B-mode harmonic imaging, Wei and colleagues have shown that the product of A×B correlates with myocardial blood flow.

**Myocardial Blood Flow Measurement**

Regional myocardial perfusion was measured by radiolabeled microsphere technology. Approximately 2 to 3 × 10^11-μm radiolabeled microspheres (New England Nuclear) were suspended in 3 mL of 0.9% saline solution/0.01% Tween 80 and injected into the left atrium during 90-second arterial reference sample withdrawal. The left ventricular slice corresponding to the MCE image was sectioned into 16 pieces that were further divided into endocardial, midwall, and epicardial portions. Samples were weighed and gamma-counted (model 5535, Packard Instruments), corrections for energy spillover into neighboring windows were made, and average transmural blood flow (mL · min⁻¹ · g⁻¹) was calculated.

**Experimental Protocol**

Three stenosis stages were modeled in each dog. A mild and a moderate non–flow-limiting stenosis and a severe flow-limiting stenosis were successively created. We defined a mild stenosis as a transstenotic gradient (difference between mean aortic pressure and LCx flow ratio) of 5 to 10 mm Hg. To create a moderate non–flow-limiting stenosis, the occluder was initially inflated until epicardial flow began to decrease to define the maximal non–flow-limiting gradient, and then partially deflated to result in a stenosis with a gradient 5 to 7 mm Hg less than the maximal gradient. To produce a flow-limiting stenosis, the occluder was inflated until a 50% to 60% reduction in epicardial flow occurred. MCE was performed at each stage, with and without adenosine (0.4 mg · kg⁻¹ · min⁻¹). Radiolabeled microsphere measurements were made during adenosine infusion at each stenosis stage and without adenosine during the moderate stenosis stage. The transstenotic gradient was continuously monitored, and MCE was performed only after at least 5 minutes of hemodynamic stability had been documented after each balloon manipulation. At the end of the experiment, the LAD was totally occluded, India ink was injected to delineate the LAD bed, the dog was euthanized, and the MCE heart slice was sectioned for radiolabeled microsphere analysis.

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With increasing stenosis, LAD hyperemic flow in response to adenosine was blunted, resulting in a progressive decrease in
the LAD/LCx flow ratios. In the absence of adenosine, flow during moderate stenosis was not significantly different from flow at baseline.

The transstenotic gradients across the LAD stenoses indicated a broad range of stenosis severity (Table 2). As expected, there was an increase in gradient with increasing stenosis, with a further increase at each stenosis during adenosine infusion. The gradient was highest during severe stenosis and did not substantially increase with adenosine, most likely owing to a decrease in flow at that stage.

**Relationship Between Power Doppler Data and Stenosis Severity**

Figure 1 illustrates an imaging sequence in a dog with a moderate non–flow-limiting stenosis before (upper panels) and during (lower panels) adenosine infusion at similar pulsing intervals. Despite the presence of a stenosis (resting transstenotic gradient 19 mm Hg), in the absence of adenosine, there was homogeneous enhancement of the LAD and LCx beds and increasing opacification at incremental pulsing intervals (Figures 1A through 1E). With adenosine, there was a contrast defect in the anteroseptal and anterior segments (region between arrows) as the LAD bed filled more slowly relative to the LCx (Figures 1G through 1I). Reflecting delayed microbubble transit, the defect was most prominent at short pulsing intervals (Figures 1G and 1H). With increasing pulsing intervals, this defect became more filled in such that the risk area was much less distinct at a pulsing interval of 99 (Figure 1J). Other notable features of the images are the absence of posterior wall attenuation typically seen in contrast-enhanced short-axis images and the dark appearance of the myocardium after microbubble destruction (Figures 1A and 1F).

Figure 2 graphs the pulsing interval versus videointensity data for the LAD and LCx beds for the dog in Figure 1. The uniform opacification of both territories before adenosine is represented by the curves in Figure 2A, which are similar for both the LAD and LCx. With adenosine (Figure 2B), there was delayed appearance of bubbles in the LAD bed compared with the LCx (prolongation of β), corresponding to the prominent defect seen at the shorter pulsing intervals in Figures 1G and 1H.

The location of the MCE perfusion defects paralleled the spatial distribution of hypoperfusion. Figure 3 maps and color codes microsphere flow in the endocardial, midwall, and epicardial layers of each of the 16 myocardial segments during adenosine infusion in the same dog with moderate stenosis shown in Figure 1. The location of the MCE perfusion defect in the LAD bed (region between arrows) corresponded to the myocardial region with reduced microsphere flow, and the endocardial/epicardial transmural flow gradient seen with radiolabeled microspheres was also visually represented on the MCE image.

Figure 4 depicts very mild and severe stenoses at increasing pulsing intervals from another dog during adenosine infusion. At baseline (Figure 4A), there was progressive myocardial enhancement in both beds with increasing pulsing interval. With severe stenosis (Figure 4C), the LCx bed filled briskly, whereas there was an LAD defect (region between arrows) that persisted even at pulsing intervals as long as 99 beats (not shown). With mild stenosis (Figure 4B), LAD bed enhancement was also delayed, but the defect was much less marked and resolved more quickly at higher pulsing intervals than with severe stenosis (Figure 4C).

The videointensity pulsing interval curves for the data shown in Figure 4 are depicted in Figure 5. There was rapid and uniform opacification of the LAD and LCx beds at baseline (Figure 5A). There was progressive prolongation of β with mild stenosis (Figure 5B), and with severe stenosis, there was delayed, incomplete filling of the LAD territory relative to the LCx (Figure 5C).

Figure 6 plots radiolabeled microsphere-derived blood flow (LAD normalized to LCx) versus the product of A×β from all 6 stages in the 9 dogs. There was a significant linear relationship between normalized LAD flow and the product of A×β derived from the curve fits to the power Doppler

### Table 2. Mean Transstenotic Gradient (mm Hg)

<table>
<thead>
<tr>
<th>Stage</th>
<th>No Adenosine</th>
<th>Adenosine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>2±1</td>
<td>6±5</td>
</tr>
<tr>
<td>Mild stenosis</td>
<td>10±4*</td>
<td>33±15†</td>
</tr>
<tr>
<td>Moderate stenosis</td>
<td>25±12*</td>
<td>39±13†</td>
</tr>
<tr>
<td>Severe stenosis</td>
<td>44±21*</td>
<td>44±11†</td>
</tr>
</tbody>
</table>

*p<0.001 vs baseline/no adenosine; †p<0.001 vs baseline/adenosine. n=9.
image data. There was a significant inverse linear relationship between stenosis severity, expressed as transstenotic gradient, and the product of $A^3B$ ($y=-0.02x+1.16, r=-0.70, P<0.001$). Furthermore, there was a significant linear relationship between the normalized (LAD/LCx) ratio of the product of $A^3B$ for the endocardium/epicardium and LAD/ LCx endocardial/epicardial flow ratios calculated with radio-labeled microspheres ($y=0.49x+0.52, r=0.60, P<0.001$).

**Discussion**

This study demonstrates for the first time that power Doppler measures of microbubble replenishment during MCE correlate with myocardial blood flow and thus can distinguish between coronary stenoses of varying severity when performed with adenosine stress. These data set the stage for the diagnosis of coronary artery disease with power Doppler MCE.

**Relationship Between Myocardial Blood Flow and Power Doppler Measurements**

The phenomenon of microbubble destruction in an ultrasound field has been the basis for an approach to the quantification of myocardial blood flow with MCE. Using B-mode imaging in a similar animal model to ours, Wei et al demonstrated that after an ultrasound pulse of sufficient power to destroy microbubbles, the rate of microbubble replenishment and the peak plateau videointensity during continuous microbubble infusion correlated with myocardial blood flow.

Whether power Doppler can generate physiologically meaningful data or reproduce the B-mode findings described by Wei et al is an important issue for several reasons. First, imaging modalities designed in theory to be advantageous over the more validated B-mode harmonics should demonstrate similar ability to detect stenoses. Second, reproduction of the previously described relationship between microbubble destruction/replenishment and blood flow by a different imaging technique would lend further support to the principles on which the Wei et al model is based and to new concepts in microcirculatory physiology emerging from this model.

It might have been predicted that power Doppler images would apply to the model derived with B-mode harmonics because the ultrasound-induced microbubble destruction and replenishment that are the basis of the model are physical events common to both techniques. On the other hand, a parallel between B-mode harmonic and power Doppler capabilities for quantifying perfusion was not inevitable for at least 3 reasons. First, with power Doppler, the amplitude of the returning Doppler signal is determined by the magnitude of variance between “packets” of ultrasound pulses. In contrast, with B-mode harmonics, the image derives from harmonic frequency components selected from a spectrum of returning frequencies representing the acoustic backscatter of bubbles and tissue. Thus, the 2 techniques register different specific features of the same acoustic event. Second, the dynamic range of power Doppler (40 dB) is less than that of B-mode harmonic imaging (60 dB) in the imaging system used in the present study. Third, the 2 techniques may differ in the efficacy of microbubble destruction. Power Doppler delivers its energy in multiple, repetitive packets of ultrasound pulses, which may inherently be more destructive than the B-mode pulse.

Given these considerations, it has not been possible to assume that power Doppler would parallel B-mode harmonics for quantifying stenoses. The major finding of the present study is that the quantitative data taken from a pure Doppler image set correlates to myocardial blood flow. In addition, as illustrated in Figures 1 and 4, qualitative visual interpretation of stenosis severity is possible, without the need for background subtraction. Moreover, these data indicate that it is possible to distinguish transmural gradients in flow from power Doppler-generated images.

Masugata and colleagues recently reported a study of dogs with flow-limiting stenoses subjected to MCE with...
power Doppler imaging, and the authors compared videointensity measures with B-mode harmonic imaging. In that study, power Doppler appeared more capable of delineating flow-limiting stenosis than B-mode imaging. The present study concurs with that study in providing data to support the capabilities of power Doppler. However, the present study differs from that of Masugata et al.22 and adds to our understanding of power Doppler MCE in several important ways. First, Masugata et al.22 evaluated flow-limiting stenoses at rest, whereas we imaged non–flow-limiting stenoses during pharmacological stress. We demonstrated the ability of power Doppler to identify even mild stenoses that are not flow limiting at rest, which more closely simulates clinically encountered scenarios. Second, we applied the exponential model of Wei et al.17 to pulsing interval versus videointensity data to derive a correlation between the MCE data and radiolabeled microsphere-derived myocardial blood flow.

Advantages of Power Doppler Imaging
Power Doppler has several theoretical advantages over B-mode harmonic imaging. Because the measurement of interest in power Doppler is the change in the returning radiofrequency signal resulting from the nonlinear behavior and/or dissolution of microbubbles in an ultrasound field, the image should, under ideal conditions, be a representation solely of the nonlinear and/or acoustic destruction events.9–11 The greater the change, the greater the amplitude calculated by the autocorrelator detector of the system. The nonlinear properties of myocardial tissue that contribute to persistent tissue background noise in B-mode harmonic imaging should be suppressed with power Doppler images. This property can be seen in Figure 1A, which demonstrates black myocardium immediately after bubble destruction.

Figure 4. MCE images from (A) baseline, (B) mild stenosis, and (C) severe stenosis stages during adenosine infusion in a second dog. Triggering intervals are indicated below each frame. Region between arrows indicates LAD bed.

The phenomenon of posterior wall attenuation in contrast-enhanced short-axis views19 has favored the use of apical imaging planes in which far-field attenuation effects are less likely to obscure the myocardium.5 This approach, however, foregoes useful segmental information imparted by short-axis views. In the present study, bright myocardial opacification was achieved at low doses of PB127, with a striking lack of posterior wall attenuation. It is likely that careful dosage selection for continuous infusion17 helped to minimize such attenuation artifact. It is also possible that properties of the agent itself, including a relatively narrow size distribution that would allow for even tighter regulation of the effective cross-sectional scattering area at a given infusion rate, contributed to the lack of attenuation in the short-axis views. An additional advantage, therefore, of the techniques used in the present study is that the lack of far-field attenuation may restore the utility of short-axis imaging planes.

Study Limitations
Because power Doppler is premised on the detection of change between successive pulses, it is susceptible to signal artifacts created by other dynamic events such as cardiac movement.11 This inherent limitation was not insurmountable, because imaging at a point in the cardiac cycle with the least cardiac movement largely overcame any significant motion artifacts at the pulse-repetition frequencies that were used.

We cannot be certain that all microbubbles were destroyed with each ultrasound pulse and therefore whether the enhancement seen at each pulsing interval represented only de novo arrival of microbubbles into the imaging plane. The appearance of dark myocardium (Figure 1), as well as our empiric confirmation of a relationship between radiolabeled microsphere flow and videointensity parameters derived from a mathematical
model that assumes complete destruction, suggests that incomplete bubble destruction was not a significant limitation. Moreover, a multipulse technique such as Doppler should have further enhanced microbubble destruction.

Generally, a high dynamic range has been advocated for contrast echo to optimize the detection of bubble signals above tissue signals, and power Doppler imaging has a lower dynamic range than B-mode harmonic imaging. Nonetheless, as indicated by our data, this property did not preclude the generation of quantifiable images. Conceivably, the impact of bubble specificity and less background tissue noise inherent to power Doppler superseded any effects of a slightly narrower dynamic range, as compared with B-mode imaging, in which a portion of the broader range is “wasted” by the background tissue signals.

We chose a gray-scale map for presentation of the power Doppler images. Subtle perfusion defects can be more difficult to appreciate because of intrinsic limitations to the visual perception of gray-level differences. Although the minimal background noise in the power Doppler images enabled visualization of defects and rendered quantifiable videointensity curves, color coding of the images, as previously described by us and others, may further facilitate visual analysis of subtle defects.

New methods for detecting microbubbles in tissue continue to evolve, including low-mechanical-index techniques that may offer the possibility of performing real-time perfusion imaging. These techniques are distinct from power Doppler, and the relative merits of one technique over the other remain to be determined.

Conclusions

We have demonstrated that detection and quantification of a range of coronary stenoses using a model based on microbubble destruction and replenishment is possible with power Doppler imaging during pharmacological stress and continuous infusion of an ultrasound contrast agent. Furthermore, excellent image quality and videodensitometric data can be achieved without the need for image processing. Minimal posterior wall attenuation in the short-axis view permits a more comprehensive analysis of segmental perfusion than is possible with the currently preferred apical views. These data provide a basis and rationale for the use of power Doppler as an approach to perfusion imaging during MCE.

Acknowledgments

Dr Villanueva is supported in part by a FIRST Award from the National Institutes of Health (R29HL58665). This study was supported in part by a grant from POINT Biomedical Corporation, which manufactures PB127, the ultrasound contrast agent used for this study.

References

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Circulation. 2001;103:2624-2630
doi: 10.1161/01.CIR.103.21.2624
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7322. Online ISSN: 1524-4539

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