Spatial Inhomogeneity of Sympathetic Nerve Function in Hibernating Myocardium

Andrew J. Luisi, Jr, MD; James A. Fallavollita, MD; Gen Suzuki, MD, PhD; John M. Canty, Jr, MD

Background—Although humans and swine with hibernating myocardium have an increased risk of sudden death, the contribution of chronic alterations in sympathetic nerve function is unknown. Acute transmural ischemia causes inhomogeneity in sympathetic innervation that may lead to lethal arrhythmias, but it is unclear whether similar abnormalities develop in response to chronic reversible ischemia.

Methods and Results—Swine were chronically instrumented with a left anterior descending coronary artery (LAD) stenosis that produced hibernating myocardium after 3 months. Resting subendocardial flow (LAD 0.75±0.14 versus 1.19±0.14 mL · min⁻¹ · g⁻¹, P<0.05) and wall thickening (LAD 15±3% versus 40±2%, P<0.05) were reduced compared with normal remote regions, without triphenyltetrazolium chloride evidence of necrosis. ¹³¹I-meta-iodobenzylguanidine (MIBG) was used to assess integrity of the norepinephrine uptake-1 mechanism, and the spatial and transmural distributions were quantified by ex vivo counting. In hibernating myocardium, MIBG deposition was decreased in each layer, with the greatest reduction in the subendocardium (LAD subendocardium 0.28±0.02 versus 0.42±0.04 mL · g⁻¹ · min⁻¹ in normal, P<0.05; LAD subepicardium 0.31±0.03 versus 0.38±0.04 mL · g⁻¹ · min⁻¹ in normal, P<0.05). In contrast, there were no spatial alterations of MIBG deposition in sham-instrumented animals.

Conclusions—The sympathetic norepinephrine uptake-1 mechanism is impaired in hibernating myocardium. These findings raise the possibility that chronic alterations in sympathetic innervation contribute to the excess mortality seen in the setting of hibernating myocardium. (Circulation. 2002;106:779-781.)

Key Words: hibernation ▪ myocardial stunning ▪ nervous system, sympathetic ▪ death, sudden ▪ ischemia

Spatial variations in regional sympathetic innervation lead to inhomogeneity in cardiac repolarization and may increase the risk of arrhythmic sudden death.¹⁻³ Regional denervation occurs after myocardial infarction, and lethal arrhythmias may be related to nerve sprouting during reinnervation.⁴ Functional denervation has also been observed after brief coronary occlusions,⁵⁻⁶ as well as in viable risk areas of reperfused myocardial infarcts,⁷ which supports the view that sympathetic nerve function is exquisitely sensitive to ischemia.

Although transmural ischemia causes cardiac denervation, it is uncertain whether reversible subendocardial ischemia could chronically affect sympathetic nerve function and contribute to the risk of sudden death in patients with chronic ischemic heart disease. In support of this, clinical studies have demonstrated an increased mortality when patients with hibernating myocardium are not revascularized, and the excess risk is largely related to sudden death.⁸ This increased mortality is also seen in animal studies of chronic hibernating myocardium, in which the cumulative incidence of sudden death approaches 50% over 5 months.⁹

We hypothesized that hibernating myocardium exhibits regional inhomogeneity in sympathetic innervation that arises from chronic repetitive ischemia distal to a critical coronary stenosis. To test this, we evaluated the functional integrity of the sympathetic nervous system by characterizing the transmural deposition of ¹³¹I-meta-iodobenzylguanidine (MIBG) in swine with chronic hibernating myocardium. The results demonstrate regional alterations in MIBG uptake, which may contribute to the pathophysiological sequelae of hibernating myocardium.
Transmural Variations in MIBG Deposition and Flow

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<th>Endo</th>
<th>Remote</th>
<th>LAD</th>
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<tbody>
<tr>
<td>MIBG deposition, mL·g⁻¹·min⁻¹</td>
<td>0.28±0.02*</td>
<td>0.42±0.04</td>
<td>0.32±0.03*</td>
<td>0.44±0.05</td>
<td>0.31±0.03*</td>
<td>0.38±0.04</td>
<td>0.93±0.05*</td>
<td>1.10±0.03</td>
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<tr>
<td>Resting flow, mL·g⁻¹·min⁻¹</td>
<td>0.75±0.14*</td>
<td>1.19±0.14</td>
<td>0.99±0.09</td>
<td>1.13±0.12</td>
<td>0.92±0.11</td>
<td>0.86±0.09</td>
<td>0.84±0.13</td>
<td>1.41±0.09</td>
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<tr>
<td>Vasodilated flow, mL·g⁻¹·min⁻¹</td>
<td>0.70±0.13*</td>
<td>4.70±0.58</td>
<td>1.56±0.33*</td>
<td>5.71±0.47</td>
<td>2.20±0.38*</td>
<td>4.81±0.33</td>
<td>0.32±0.04*</td>
<td>0.97±0.07</td>
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<tr>
<td>Flow reserve</td>
<td>1.10±0.24*</td>
<td>4.48±0.87</td>
<td>1.63±0.34*</td>
<td>5.67±0.95</td>
<td>2.61±0.46*</td>
<td>6.26±1.02</td>
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</table>

Endo indicates subendocardiun; Epi, subepicardium. Mean±SEM.
*P<0.05, LAD vs. Remote.

confused to maintain mean arterial pressure. After measurements were completed, catheters were removed, and the animals recovered uneventfully.

To circumvent potential effects of adenosine and catecholamine stimulation on sympathetic function, we assessed the regional distribution of MIBG in the closed-chest sedated state 5±1 days later. After hemodynamics had equilibrated, MIBG (100 to 250 μCi IV) was injected and an arterial withdrawal sample obtained to integrate the time-activity curve. After 45 minutes, animals were deeply anesthetized (isoflurane) and hearts removed for sampling. The left ventricle was cut into 4 circumferential rings, and each ring was divided into ~12 wedges. Wedges were subdivided into 3 transmural layers. Intervening rings were stained with triphenyltetrazolium chloride to exclude infarction. Samples were weighed, and decay-corrected activity was determined by counting in a sodium-iodide detector (Wallac, Inc.). MIBG deposition was determined by dividing sample activity by the integrated arterial activity. After activity was allowed to decay to background, samples were digested and processed for microsphere flow analysis as described previously. Data are expressed as mean±SEM, with the P<0.05 level considered significant.

### Results

At the time of study, heart rate averaged 94±7 bpm, left ventricular end-diastolic pressure averaged 23.3±1.6 mm Hg, and aortic systolic and diastolic pressures averaged 147±7 and 89±4 mm Hg, respectively. Hibernating myocardium demonstrated anteroseptal hypokinesis (wall thickening 15±3% versus 40±2% in remote regions, P<0.05), with no triphenyltetrazolium chloride evidence of infarction. Measurements of flow are summarized in the Table.

Resting flow was reduced in hibernating myocardium (LAD subendocardial flow 0.75±0.14 versus 1.19±0.14 mL·g⁻¹·min⁻¹, P<0.05), and subendocardial flow during adenosine was unable to increase above the resting level.

There was spatial inhomogeneity in the distribution of MIBG that which varied significantly by region and by myocardial layer (2-way ANOVA, P<0.05). Figure 1 summarizes the circumferential distribution of MIBG in full-thickness samples. MIBG was systematically reduced in hibernating LAD regions (0.30±0.02 versus 0.41±0.05 mL·g⁻¹·min⁻¹ in remote regions), and the pattern of reduced MIBG paralleled the area of reduced vasodilated flow. Reductions in MIBG were similar in stenotic (n=2) and totally occluded, collateral-dependent animals (n=6). These spatial variations did not arise from dissection of the coronary artery because sham animals demonstrated no regional alterations in MIBG.

Figure 2 summarizes transmural variations in MIBG deposition. Relative deposition (LAD/remote) was significantly reduced in each myocardial layer, with the greatest reduction in the subendocardium (0.68±0.04 versus 0.81±0.03 in the subepicardium, P<0.05). Subendocardial MIBG deposition in hibernating myocardium was also lower than corresponding values in the subepicardium (endocardium/epicardium MIBG 0.93±0.05 versus 1.10±0.03 in remote regions, P<0.05). Nevertheless, the transmural gradient for MIBG was not as steep as that for adenosine perfusion (endocardium/epicardium adenosine flow; LAD 0.32±0.04 versus 0.97±0.07 in remote regions, P<0.05).

### Discussion

The major new finding from the present study is that hibernating myocardium exhibits chronic spatial inhomogeneity in the norepinephrine uptake-1 mechanism, as reflected by reductions in MIBG deposition. Although these changes are similar to those reported in viable myocardium after transmural infarction, they occur in the absence of myocardial necrosis and are most pronounced in the
subendocardium. This raises the possibility that inhomogeneity of sympathetic innervation arising from reversible ischemia may contribute to the increased risk of sudden cardiac death when hibernating myocardium cannot be revascularized.8

Approximately 50% of myocardial MIBG deposition is dependent on the presynaptic norepinephrine uptake-1 mechanism. The remainder is nonspecific uptake and not affected by denervation.15 Although controversy exists as to whether acute reductions in MIBG uptake reflect the uptake-1 mechanism after acute infarction,13–15 β-adrenergic–mediated changes in myocardial function3 and α-adrenergic coronary vasoconstriction6 in response to sympathetic nerve stimulation are immediately attenuated after brief reversible episodes of transmural ischemia. In addition, animal models have shown that reductions in MIBG activity are associated with reductions in tissue norepinephrine content and sympathetic nerve density, which supports the use of MIBG as an index of sympathetic nerve function in the chronic setting.13

The circumferential reduction in MIBG uptake in hibernating myocardium paralleled the area of chronically reduced flow reserve and is consonant with the observation that sympathetic denervation corresponds to the area at risk of ischemia in ST-elevation myocardial infarcts.7 This supports the notion that the propensity of a region to develop spontaneous ischemia is closely linked to the development of sympathetic denervation. There was also a modest transmural gradient in MIBG deposition, with greater reductions found in the subendocardium, where flow reserve was critically impaired. Compared with normal remote regions, the 32% relative reduction in MIBG that we found in the subendocardium of hibernating myocardium exceeded the ~18% difference noted in the subendocardium of dysfunctional, normally perfused myocardium adjacent to transmural infarction.14 In contrast, the relative differences in subepicardial MIBG were similar (19% and 17%, respectively). The reduction in subepicardial MIBG could reflect an increased sensitivity of sympathetic nerves to even modest reductions in flow reserve and transient demand-induced ischemia. An alternate possibility is that they arise from transient total coronary occlusion and supply-induced ischemia from cyclic platelet aggregation before the development of total coronary occlusion, as previously described in amiodarone occluder models.16 Finally, it is also possible that factors unrelated to ischemia could contribute to subepicardial denervation, as they do in normally perfused border zones adjacent to transmural infarction.14

Clinical Implications
Our data raise the possibility that regional inhomogeneity in sympathetic nerve function may be one of the determinants of increased mortality from sudden death in patients with hibernating myocardium. This is also consistent with the high rate of sudden death in the swine model of hibernating myocardium,9 which we have demonstrated to be associated with the development of ventricular tachycardia that degenerates rapidly into ventricular fibrillation (unpublished observations). Although additional studies are necessary, imaging the integrity of the sympathetic nervous system may provide an approach to stratify risk of sudden death in patients with hibernating myocardium and depressed left ventricular function.

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References
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