Moderate Systemic Hypotension During Reperfusion Reduces the Coronary Blood Flow and Increases the Size of Myocardial Infarction in Pigs*

John N. Nanas, MD; Elias Tsolakis, MD; John V. Terrovitis, MD; Ageliki Eleftheriou, MD; Stavros G. Drakos, MD; Argirios Dalianis, MD; and Christos E. Charitos, MD

Study objectives: To examine the effects of low arterial BP (ABP) during reperfusion on the extent of myocardial infarction and on coronary blood flow (CBF) in an occlusion/reperfusion experimental model.

Design: Prospective, randomized animal study.

Setting: University hospital.

Participants: Normal pigs that were anesthetized, intubated, and mechanically ventilated.

Interventions: Twenty-seven open-chest pigs underwent occlusion of the mid left anterior descending (LAD) coronary artery for 1 h followed by reperfusion for 2 h. During reperfusion, the animals were randomly assigned to either continuous infusion of nitroglycerin in therapeutic doses and fluid infusion at rates to maintain a mean ABP ≥ 80 mm Hg (group 1, n = 13), or continuous nitroglycerin infusion at rates to maintain a mean ABP between 60 mm Hg and 75 mm Hg (group 2, n = 14).

Measurements and results: The hemodynamics and the coronary ABP distal to the occlusion were recorded throughout the experiment. In addition, the LAD CBF and peak hyperemia CBF before occlusion and during reperfusion periods were measured by transit-time flowmetry. At the end of the experiment, the infarcted left ventricular myocardial size was measured. There were no significant hemodynamic differences, including the distal coronary arterial pressure, between the two groups before or during the LAD artery occlusion period. During reperfusion, mean ABP was 90 ± 3 mm Hg in group 1 vs 69 ± 3 mm Hg in group 2 (p < 0.001). In group 1, the infarcted myocardium represented 50.3 ± 4.3% of the myocardium at risk, vs 69.4 ± 7.2% in group 2 (p < 0.001). During reperfusion, CBF and peak hyperemia CBF were significantly higher in group 1 than in group 2.

Conclusions: Low ABP during reperfusion increases the size of myocardial infarction and decreases CBF.

Key words: blood flow; BP; collateral circulation; infarction; reperfusion

Abbreviations: ABP = arterial BP; CBF = coronary blood flow; LAD = left anterior descending; LV = left ventricular; MR = myocardium at risk; DCAP = distal coronary arterial pressure

Experimental studies1,2 have shown that infarct size and rate of infarct progression in dogs with chronic hypertension and left ventricular (LV) hypertrophy are markedly influenced by the level of LV systolic pressure during coronary arterial occlusion, while normotension induced by renal anastomosis or nitroprusside during myocardial ischemia resulted in infarct size similar to normotensive dogs without LV hypertrophy. However, in observations made in the thrombolytic era, a systolic arterial BP (ABP) < 120 mm Hg in patients with acute myocardial infarction was associated with a higher mortality than in patients with ABP > 120 mm Hg. Surprisingly, an ABP even higher than 160 mm Hg did not influence survival.3

*From the University of Athens School of Medicine, Department of Clinical Therapeutics, “Alexandra” Hospital, Athens, Greece. Manuscript received March 5, 2003; revision accepted August 29, 2003.

Reproduction of this article is prohibited without written permission from the American College of Chest Physicians (e-mail: permissions@chestnet.org).

Correspondence to: John N. Nanas, MD, Makedonias 24, 104 33 Athens, Greece; e-mail: Jnanas@ath.forthnet.gr
In experimental studies using a canine model, the administration of various pharmacologic agents that reduced the ABP during coronary artery occlusion with or without reperfusion showed conflicting effects on the extent of myocardial infarction. These differences could be attributed to the proved cytoprotective effect of those agents that reduced the infarct size and to the absence of such effect in those that failed to eliminate the infarct size. The IV infusion of nitroglycerin in dogs after coronary artery ligation in doses that reduced the BP by more than 10% of the baseline value was not associated with reduced infarct size. However, in that experimental model, nitroglycerin in doses that decreased the BP by a mean of 10% of the baseline value increased collateral blood flow and decreased the extent of myocardial infarction. Neither experimental nor clinical data are available regarding the effects of a decreased ABP during reperfusion on the extent of myocardial infarction. The purpose of this study was to examine the effects of lowering the systemic BP during reperfusion only, on myocardial infarct size and coronary blood flow (CBF) in an occlusion/reperfusion porcine model.

**Materials and Methods**

**Surgical Preparation**

All animals used in this study received humane care in compliance with the guidelines for care and use of laboratory animals of the US National Institutes of Health. Twenty-seven pigs weighing 22 to 35 kg were premedicated with ketamine hydrochloride (15 mg/kg IM) and midazolam (0.5 mg/kg IM), anesthetized with thiopental sodium (9 mg/kg IV bolus) and fentanyl citrate (0.5 mg IV bolus), followed by continuous IV infusions of thiopental sodium (1 mg/min), fentanyl citrate (4 mg/min), pancuronium bromide (0.25 mg/min), and lidocaine (2 mg/min), throughout the experiment. After intubation, respiration was controlled with a Soxtronic volume respirator (Soxil S.P.A.; Segrate, Italy), supplying a mixture of room air and oxygen. Tidal volume, respiratory rate, and percentage of inspiration were adjusted to maintain an arterial pH between 7.35 and 7.45, P_{CO_2} = 35 mm Hg, and P_{O_2} > 100 mm Hg. The chest was opened via a midline sternotomy, and the heart was suspended in a pericardial cradle. Catheters were placed in the aortic arch via the right carotid artery, in the right atrium via the right external jugular vein, and in the left atrium directly through the left atrial appendix. The proximal to mid left anterior descending (LAD) coronary artery was dissected free and instrumented from proximal to distal with an ultrathin catheter (0.018 inches) for infusion of adenosine, a transit time flowmeter probe, a loose ligature and, distally, an ultrathin catheter to measure distal coronary artery pressure (DCAP). The temperature was kept within 0.5 °C of the baseline value. ECG, for severe rhythm disturbances, and arterial pressure were continuously monitored. No pharmacologic agents other than anesthetics, lidocaine, sodium bicarbonate, nitroglycerin, heparin, and normal saline solution were infused. However, in the experiments in which the CBF was recorded, in order to achieve peak hyperemia CBF, on each occasion (eight times during the experiment), intracoronary adenosine (40 μg/kg/min) was infused for approximately 2 min. All experimental animals underwent occlusion of the LAD for 1 h followed by reperfusion for 2 h.

**Physiologic Measurements**

After the completion of the preparation, a few minutes were allowed for hemodynamic stabilization before the baseline recordings of aortic and right atrial pressures, DCAP, LAD CBF, and peak hyperemia CBF. Subsequently, pressures were recorded immediately after the occlusion, and every 15 min during the 1-h period of occlusion. The CBF was recorded at the second minute after the onset of reperfusion and thereafter at 5, 15, 30, 60, 90, and 120 min of reperfusion, along with peak hyperemia CBF and pressures.

**Experimental Protocol**

The LAD artery was then occluded for 1 h when the snare was released, and reperfusion was established for 2 h. The animals were excluded from the study if the baseline mean ABP was < 75 mm Hg. Two groups were studied. In group 1 (n = 13), IV nitroglycerin was infused during the reperfusion period at a rate of 2.5 μg/min, along with a crystalloid solution titrated to maintain a mean ABP ≥ 80 mm Hg. In group 2 (n = 14), IV nitroglycerin was infused during the reperfusion period at a mean rate of 100 ± 87 μg/min to decrease the mean ABP by > 20% of baseline, and to maintain it between 60 mm Hg and 75 mm Hg. Collateral circulation was monitored during the occlusion period by measuring peripheral coronary arterial pressure in seven group 1 and eight group 2 experiments. In the remaining experiments (n = 6 in each group), LAD CBF was measured. The peak hyperemia CBF was obtained after a transient 10 s of coronary occlusion, during a plateau of hyperemia achieved with intracoronary adenosine infusion at a rate of 40 μg/kg/min, and corresponded to the higher CBF obtained with these interventions.

Three hours after the initial occlusion, the LAD coronary artery was reoccluded and gentian violet (1%, 3 mL/kg) was injected into the left atrium over 30 s. Within the next 5 s, the heart was electrically fibrillated and immediately excised.

**Morphometric Measurements**

The left ventricle, including the septum, was separated from the remainder of the heart and cut into 1-cm-thick sections perpendicular to the apex-base axis. The LV area at risk of infarction was identified by the absence of gentian violet dye. The borders of the area at risk were traced with sinus dye on the heart slices. All sections were placed into a 1% triphenyltetrazolium chloride solution at 37°C for 20 min, allowing the identification of the infarcted area, which remains unstained due to lack of nicotinamide-adenine dinucleotide or substrate stores. Tracings of both sides of each ventricular section, as outlined by the gentian violet and the triphenyltetrazolium chloride reaction, were drawn on transparent plastic sheets, and the areas of each of the demarcated regions of each ventricular section underwent planimetry.

All slices were weighed. For each experiment, the myocardium at risk (MR) was calculated as a percentage of the whole left ventricle; the infarcted myocardium, including the hemorrhagic myocardium, was calculated as a percentage of the MR, as previously described in detail. The morphometric measurements, including the tracing, planimetry, and final calculations, were performed by an investigator unaware of the experimental assignment.
Statistical Analysis

The LAD CBF and the peak hyperemia CBF were each normalized to their baseline values. Data for each group were expressed as mean ± SD. Unpaired Student t test was used to examine differences between the two experimental groups with respect to hemodynamic measurements, size of infarcted myocardium, and size of MR. Repeat measurements analysis was used to evaluate differences between hemodynamic results within each experimental group at baseline, during coronary occlusion, and during reperfusion.

RESULTS

Hemodynamics

The hemodynamic measurements at baseline and during the 1-h period of coronary occlusion were similar in the two groups (Table 1). DCAP was identical to the mean aortic pressure before occlusion of the coronary artery and during the reperfusion period in both groups. During the 1 h of LAD artery occlusion, mean DCAP was 17.4 ± 3.2 mm Hg in group 1 vs 17.9 ± 4.7 mm Hg in group 2 (p = 0.721) and mean coronary arterial perfusion pressure (DCAP − right atrial pressure) was 9.7 ± 2.5 mm Hg in group 1 vs 9.4 ± 3.0 mm Hg in group 2 (p = 0.644).

During reperfusion, mean aortic pressure and double product (systolic BP × heart rate), an index of the afterload, were significantly higher in group 1 than in group 2: 90 ± 3 mm Hg vs 69 ± 3 mm Hg (p < 0.001) and 14,648 ± 657 mm Hg/min vs 11,256 ± 504 mm Hg/min (p < 0.001). The baseline CBF was 24 ± 9 mL/min in group 1 and 30 ± 20 mL/min in group 2 (p = 0.509). In group 1, a high CBF was measured during the first 5 min of reperfusion, and gradually decreased though remained above baseline and significantly higher than in group 2 throughout the 2 h of reperfusion (Fig 1). In group 2, CBF increased significantly above baseline at the onset of reperfusion, though it returned to baseline levels at 15 min, and continued to decrease gradually thereafter during the 2 h of reperfusion (Fig 1). In group 1, peak hyperemia CBF was moderately lower at 5 min after the onset of reperfusion than at baseline, and remained stable thereafter throughout the experiment (Fig 2). In group 2, peak hyperemia CBF at 5 min after the onset of reperfusion was markedly decreased to less than half of the baseline level and, after a significant further fall, reached a plateau at 15 min of reperfusion (Fig 2). During the 2 h of reperfusion, peak hyperemia CBF was significantly lower than at baseline in both groups and, in group 1, remained significantly higher than in group 2 (Fig 2).

Morphometric Results

The amount of infarcted myocardium, including the hemorrhagic myocardium, was significantly

<table>
<thead>
<tr>
<th>Group 1</th>
<th>Group 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart Rate, beats/min</td>
<td>79.8 77.4</td>
</tr>
<tr>
<td>Mean Arterial Pressure, mm Hg</td>
<td>115.9 13.6</td>
</tr>
<tr>
<td>Systolic Arterial Pressure, mm Hg</td>
<td>159.4 17.1</td>
</tr>
<tr>
<td>Diastolic Arterial Pressure, mm Hg</td>
<td>111.7 14.1</td>
</tr>
<tr>
<td>Pressure Product, mm Hg × 1,000</td>
<td>16.8 15.6</td>
</tr>
<tr>
<td>Baseline</td>
<td>120.0 10.3</td>
</tr>
<tr>
<td>15-min occlusion</td>
<td>127.3 18.1</td>
</tr>
<tr>
<td>30-min occlusion</td>
<td>122.6 16.5</td>
</tr>
<tr>
<td>60-min occlusion</td>
<td>132.9 20.9</td>
</tr>
<tr>
<td>90-min occlusion</td>
<td>136.4 22.8</td>
</tr>
<tr>
<td>120-min occlusion</td>
<td>138.5 23.1</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Variables</th>
<th>Group 1</th>
<th>Group 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>110.6 17.1</td>
<td>114.4 17.1</td>
</tr>
<tr>
<td>15-min occlusion</td>
<td>122.6 24.7</td>
<td>122.6 24.7</td>
</tr>
<tr>
<td>30-min occlusion</td>
<td>119.5 16.5</td>
<td>120.0 16.5</td>
</tr>
<tr>
<td>60-min occlusion</td>
<td>132.9 20.9</td>
<td>132.9 20.9</td>
</tr>
<tr>
<td>90-min occlusion</td>
<td>136.4 22.8</td>
<td>136.4 22.8</td>
</tr>
<tr>
<td>120-min occlusion</td>
<td>138.5 23.1</td>
<td>138.5 23.1</td>
</tr>
</tbody>
</table>

Data are presented as mean ± SD. *p < 0.05 compared with group 2 at the same time point.
smaller in group 1 (50.3 ± 4.3%) than in group 2 (69.4 ± 7.2%, p < 0.001; Fig 3). This difference remained significant when including the intracoronary adenosine infusion experiments (54.5 ± 12.8% and 64.8 ± 11.7%, respectively [p = 0.047]). The extent of MR, expressed as percentage of the LV myocardium, was similar in both groups (26.8 ± 9.6% vs 27.3 ± 10.3% of the LV myocardium in group 1 vs group 2, p = 0.940).

**DISCUSSION**

**Experimental Animals and Collateral Circulation**

The pigs used as experimental models in the present study are considered to have no, or only minimal, collateral flows on abrupt occlusion of a coronary artery. Our recording of a low peripheral coronary arterial perfusion pressure in both groups of animals was consistent with the presence of a negligible collateral circulation in this model, as has been described by other investigators.

**Nitroglycerin and Extent of Myocardial Infarction**

In the thrombolytic era, large clinical trials of nitrate therapy in acute myocardial infarction found no independent effect of nitrates on mortality, suggesting that nitroglycerin has no direct effect on infarct size when administered during reperfusion. In this study, IV infusion of nitroglycerin was used as an experimental tool to decrease the ABP during reperfusion in group 2; in the absence of experimental confirmation of the above hypothesis, our study design included the infusion of nitroglycerin during reperfusion in both experimental groups, in therapeutic doses in group 1 and in moderately hypotensive doses in group 2.

**Adenosine and Extent of Myocardial Infarction**

In this study, the peak hyperemia was evaluated during reperfusion after 1 h of ischemia; therefore, the coronary vascular function was severely impaired. In order to ascertain the achievement of maximum coronary vasodilation, the peak hyperemia was obtained after transient ischemia during intracoronary adenosine infusion. Adenosine induces preconditioning and reduces infarct size if administered before coronary artery occlusion, and inhibits neutrophil-mediated endothelial cell injury, reduces infarct size, prevents the no-reflow phenomenon, and improves ventricular function if administered at the time of reperfusion. It is possible that the infusion of adenosine before arterial occlusion prolonged the duration of occlusion necessary to cause infarction, and that its repeated infusion during reperfusion may have mitigated reperfusion injury and limited infarct size.

In the present study, the extent of myocardial...
Infarction was significantly smaller in group 1 than in group 2, with and without inclusion of the experiments in which adenosine was used. The differences in infarct size between animals with high normal levels (group 1) vs animals with low normal levels (group 2) of arterial pressure during reperfusion seems likely to be due to a negative effect of the lower ABP on the extent of myocardial infarction.

ABP

The present study is the first to prospectively examine the effects of moderately reduced aortic pressure during the myocardial reperfusion on the extent of myocardial infarction. Neither experimental nor clinical data are available regarding the effects of a decreased ABP during reperfusion on the extent of myocardial infarction. However, increasing the diastolic aortic pressure during reperfusion in normotensive dogs reduced the size of myocardial infarction. The level of ABP during reperfusion may influence the extent of the reperfusion injury.

Reperfusion Injury and Postischemic CBF

Experimentally and clinically, reperfusion after severe myocardial ischemia or infarction precipitates postreperfusion hemorrhage. In this study, the hemorrhagic component of myocardial infarction was a fraction of the whole calculated area of infarcted myocardium. Another factor known to limit the beneficial effects of blood flow restoration after ischemia is the inability of myocardial tissue to be reperfused. It is well established that blood flow distribution to previously ischemic myocardium is heterogeneous and may be lower than flow to the nonischemic region. This multifactorial “no-reflow” phenomenon is the consequence of severe ischemic damage to the microvasculature, resulting in its obstruction at the moment of reperfusion from capillary compression by myocardial edema, myocardial ischemic contracture, direct ischemic microvascular injury with endothelial cell swelling and an increased vasomotor tone. Microvascular obstruction by erythrocyte and leukocyte plugging or platelet aggregation also seems to participate in the development of no-reflow, which influences infarct size and CBF during reperfusion.

In our study, the CBF and peak hyperemia CBF during reperfusion were lower in the animals with a decreased ABP than in animals with a stable ABP during reperfusion. The decrease in CBF below baseline values after a brief initial increase in group 2, in contrast to the gradual decrease and maintenance to above baseline values throughout the 2 h of reperfusion in group 1, is particularly noteworthy. These serial changes in blood flow in postischemic myocardium are consistent with those observed in previous studies. Using radioactive microspheres in pigs undergoing 40 min of coronary artery occlusion, Johnson et al found a reduction in peak CBF at 60
min of reperfusion comparable to that measured in group 2 of our study; likewise, infarct size was similar to that of our group 2. It is also noteworthy that the systolic BP during reperfusion was unintentionally decreased from a mean baseline value of 120 mm Hg to 90 mm Hg, a decrease commensurate to that measured during reperfusion in our group 2. In addition, a close correlation ($r = -0.79$) was found between the extent of infarction and peak hyperemia CBF. In dogs, Cobb et al$^{31}$ found the CBF to be hyperemic in all myocardial layers during the first 15 min of reperfusion after prolonged coronary occlusion, and to be significantly decreased at 4 h in regions with $>50\%$ infarction. Reffelmann et al$^{32}$ found a significant reduction in myocardial blood flow between 5 min and 2 h of reperfusion. Furthermore, in isolated rat hearts subjected to 30 min of no-reflow ischemia, a reduction in coronary flow during reperfusion enhanced the trapping of leukocytes in capillaries, and adhesion of leukocytes in venules was inversely related to shear rate.$^{33}$ Based on these earlier data, we hypothesize that during reperfusion, low normal BP is associated with decreased CBF because it cannot provide adequate reperfusion in myocardial regions with severely increased coronary vascular resistance. This decreased CBF accelerates leukocyte accumulation, enhances the no-reflow phenomenon, and increases the extent of myocardial infarction in group 2. This hypothesis finds support in the observation of a decrease in ABP, but not CBF, by prostaglandin I$_2$ during the reperfusion period, most likely caused by inhibition of neutrophil function.$^{5,7}$ A delayed, progressive fall in CBF in areas that initially were properly reperfused has been observed in regions receiving no collateral flow during coronary occlusion, which is associated with accumulation of neutrophils and plugging of the capillaries late in the course of reperfusion. In that study, CBF was markedly decreased within the first 30 min of reperfusion with a further slight reduction at 3.5 h of reperfusion.$^{34}$

CBF during reperfusion in both our experimental groups gradually decreased over time, though this was more prominent in the group with a low ABP during reperfusion. Since the animals had no evidence of collateral blood flow during occlusion of the LAD artery, which predisposed them to the development of no-reflow, the pattern of CBF during reperfusion was probably related to the accumulation of neutrophils, apparently enhanced by the lower CBF in group 2.$^{34}$

**Clinical Implications**

The harmful effects on infarct size of a low normal ABP during reperfusion observed in this experimen-
tal study suggest that it should be avoided in a clinical situation. Reperfusion during acute myocardial infarction is usually achieved by thrombolysis or mechanical vessel dilatation. In normotensive patients after such interventions, a low normal ABP in the absence of other signs of hemodynamic instability can be increased to high normal levels by the infusion of crystalloid solutions. The results of this study support the idea that post-myocardial infarct preehox, defined as persistent low normal ABP in the presence of other signs of hemodynamic instability, including sinus tachycardia and a third heart sound, should be treated with the intra-aortic balloon pump, which augments the diastolic aortic pressure, decreases myocardial infarct size, and increases survival.

**CONCLUSION**

Our experiments showed that in normotensive animals, a decreased central ABP during myocardial reperfusion, even when kept within normal limits, is associated with a larger myocardial infarct size and lower levels of CBF. This suggests that in normotensive patients a decreased ABP, in the setting of acute myocardial infarction in the absence of other signs of hemodynamic instability, must be corrected to high normal levels. Previous experimental and clinical observations, combined with the findings of the present study, allow the formulation of a hypothesis, to be tested in a randomized clinical study of patients undergoing coronary reperfusion in the setting of acute myocardial infarction.

**REFERENCES**

15. GISSI-3: effects of lisinopril and transdermal glyceryl trinitrate singly and together on 6-week mortality and ventricular function after acute myocardial infarction. Lancet 1994; 343:1115–1122