Synergism Between Platelets and Neutrophils in Provoking Cardiac Dysfunction After Ischemia and Reperfusion
Role of Selectins

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Background—Neutrophils (PMNs) are known to contribute to both cardiac dysfunction and myocardial necrosis after reperfusion of an ischemic heart. Moreover, platelets are also important blood cells that can aggravate myocardial ischemic injury. This study was designed to test the effects of PMNs and platelets separately and together in provoking cardiac dysfunction in isolated perfused rat hearts after ischemia and reperfusion.

Methods and Results—Control rat hearts not subjected to ischemia were perfused without blood cells for 80 minutes. Additional control rat hearts were perfused with 75×10^6 PMNs, with 100×10^6 platelets, or with 75×10^6 PMNs + 100×10^6 platelets over a 5-minute perfusion followed by a 75-minute observation period. No significant reduction in coronary flow, left ventricular developed pressure (LVDP), or the first derivative of LVDP (dP/dt max ) was observed at the end of the observation period in any nonischemic group. Similarly, global ischemia (I) for 20 minutes followed by 45 minutes of reperfusion (R) produced no sustained effects on the final recovery of any of these parameters in any group of hearts perfused in the absence of blood cells. However, I/R hearts perfused with either PMNs or platelets alone exhibited decreases in these variables of 10% to 12% (P<0.05 from control). Furthermore, I/R hearts perfused with both PMNs and platelets exhibited decreases of 50% to 60% in all measurements of cardiac function (P<0.001). These dual-cell–perfused I/R hearts also exhibited marked increases in cardiac myeloperoxidase (MPO) activity, indicating a significant PMN infiltration, and enhanced P-selectin expression on the coronary microvascular endothelium. All cardiodynamic effects as well as MPO accumulation and PMN infiltration were markedly attenuated by a sialyl LewisX-oligosaccharide or a recombinant soluble P-selectin ligand, which inhibits selectin-mediated cell adhesion.

Conclusions—These results provide evidence that platelets and neutrophils act synergistically in provoking postreperfusion cardiac dysfunction and that this may be largely due to cell-to-cell interactions mediated by P-selectin. These findings may help explain the reperfusion injury phenomenon. (Circulation. 1998;98:1322-1328.)

Key Words: myocardium ■ cell adhesion molecules ■ contractility ■ selectins

Reperfusion of the ischemic myocardium often results in neutrophil (PMN)-induced myocardial injury as well as in cardiac contractile dysfunction.1–5 PMNs accumulate in the microvasculature, which can lead to microvascular plugging, resulting in impaired coronary perfusion.4,5 Postreperfusion cardiac necrosis and cardiac contractile dysfunction can be prevented by depletion of circulating leukocytes6 or by monoclonal antibodies and ligand blockers directed against cell adhesion molecules.3,9 More recently, exogenous administration of physiological levels of a nitric oxide donor7,11 has been found to markedly attenuate the cardiac dysfunction induced by PMNs in perfused rat hearts subjected to ischemia and reperfusion. In these isolated perfused rat heart studies, PMNs were added to plasma, and this mixture was added to the Krebs-Henseleit perfusion fluid.10,11 However, there were no platelets in the cardiac perfuse in these experiments.

Platelets are a known source of a variety of inflammatory mediators, including thromboxane A_2,12 P-selectin,13 and platelet factor 4,14 all of which upregulate either the complement system or a variety of cytokines.14,15 PMNs also release a variety of cell-activating and cytotoxic mediators, including cytokines (ie, tumor necrosis factor-α, interleukin-1β), complement products (eg, C5a6–9), proteolytic enzymes (eg, elastase, cathepsin G), and oxygen-derived free radicals (eg, superoxide) that may contribute to platelet activation.16–18 However, little is known regarding cross-talk between PMNs and platelets in contributing to the pathophysiology of myocardial ischemia and related cardiovascular disease states. Recently, Neumann et al15 showed that leukocyte-platelet binding is increased in patients with acute myocardial infarction as well as in patients with unstable angina.19 More recently, others have shown that platelets and PMNs interact...
in regulating vascular tone in arteries injured by angioplasty.\textsuperscript{20} Nevertheless, the nature of the interaction between platelets and PMNs is not known with regard to their influence on cardiac performance in myocardial ischemia/reperfusion injury or how these 2 blood cell types communicate with the vasculature.

The major purpose of this study was to investigate the effect of platelets and PMNs separately and together on cardiac performance in a well-controlled, established model of ischemia/reperfusion in the isolated perfused rat heart and to assess the role of the selectin family of glycoprotein adhesion molecules in modulating cardiac dysfunction in blood–perfused rat hearts subjected to myocardial ischemia/reperfusion.

Methods

**Isolated Rat Heart Experiments**

Male Sprague-Dawley rats (250 to 300 g) were anesthetized with 40 mg/kg sodium pentobarbital and given 1000 U sodium heparin IP (Abbott Laboratories, Diagnostic Division). After a midline thoracotomy, the hearts were rapidly excised, the ascending aorta was cannulated, and retrograde perfusion of the heart was initiated on a Langendorff apparatus at a constant pressure of 80 mm Hg. These isolated hearts were perfused with a Krebs bicarbonate buffer of the following composition (in mmol/L): glucose 17, NaCl 120, NaHCO\textsubscript{3} 25, CaCl\textsubscript{2} 2.5, EDTA 0.5, KCl 5.9, and MgCl\textsubscript{2} 1.2, maintained at 37°C. The perfusate was oxygenated with 95% O\textsubscript{2}/5% CO\textsubscript{2}, which equilibrated at a pH of 7.3 to 7.4. Two sidearms in the perfusion line located just proximal to the heart inflow cannula allowed infusion of PMNs, platelets, and plasma directly into the coronary inflow line.

To assess cardiac contractile function, a 2.5F microtip catheter located just proximal to the heart inflow cannula allowed infusion of PMNs, platelets, and plasma directly into the coronary inflow line. To assess cardiac contractile function, a 2.5F microtip catheter located just proximal to the heart inflow cannula allowed infusion of PMNs, platelets, and plasma directly into the coronary inflow line. To assess cardiac contractile function, a 2.5F microtip catheter located just proximal to the heart inflow cannula allowed infusion of PMNs, platelets, and plasma directly into the coronary inflow line.

**Rat Neutrophil Isolation**

Neutrophil donor rats (300 to 350 g) received a 10-mL injection of 0.5% glycogen IP (Sigma Chemical Co). Eighteen hours later, the rats were anesthetized with ethyl ether, and the neutrophils were harvested by peritoneal lavage in PBS. The peritoneal lavage was centrifuged at 3000 rpm for 10 minutes, and the plasma was decanted from the blood cell–perfused rat hearts subjected to myocardial ischemia/reperfusion.

**Rat Platelets and Plasma**

Whole blood was obtained by an intracardiac puncture in anesthetized rats with a 20-mL plastic syringe with a 20-gauge needle (Becton Dickinson) containing 2000 U sodium heparin. To obtain platelets, the whole blood was immediately spun in a refrigerated centrifuge (GSGR; Beckman Instruments, Inc) at 900 rpm for 10 minutes. Theuffy coat was collected and centrifuged again at 3000 rpm for 10 minutes, and the plasma was decanted from the platelets. Next, the platelets were resuspended in Krebs buffer and counted. Platelet preparations were >95% pure.

**Perfused Heart Experimental Protocol**

After a 15-minute stabilization period, baseline left ventricular developed pressure (LVDP), +dP/dt\textsubscript{max}, and coronary flow were measured every 5 minutes for 15 minutes to ensure complete equilibration of the hearts. Flow of Krebs buffer was then reduced to zero, creating global, total ischemia. This ischemia was maintained for 20 minutes. Reperfusion of the hearts was instituted by restoration of flow of buffer to the heart to that of preischemic levels. At 5 minutes of reperfusion, either 75×10\textsuperscript{6} PMNs alone, 100×10\textsuperscript{6} platelets alone, or both cell types together were infused into the hearts. In a separate series of 5 rat hearts, 75×10\textsuperscript{6} platelets plus 750×10\textsuperscript{6} platelets were infused together in rat hearts subjected to the same ischemia-reperfusion protocol. A few additional hearts were perfused with either 100×10\textsuperscript{6} PMNs or 50×10\textsuperscript{6} platelets. The blood cells were infused over a 5-minute period directly into the coronary circulation via a set of side ports situated in the perfusion line just proximal to the inflow cannula. The PMNs were suspended in 5.0 mL of Krebs buffer in a 5.0-mL syringe, and the platelets were suspended in 5.0 mL of plasma and also placed in a separate 5.0-mL syringe located just distal to the platelet inflow port and just proximal to the inflow port to the coronary circulation. The hearts were allowed to reperfuse for a total of 45 minutes, during which time data were collected every 5 minutes for the first 30 minutes and at the 45-minute time point. A sialyl Lewis\textsuperscript{a}-oligosaccharide (SLe\textsuperscript{a}-OS) (CY-1503) was obtained from Cytel, Inc and diluted in Krebs buffer.\textsuperscript{21} In additional experiments, we used a soluble P-selectin ligand (sPSGL-1) described by Takada et al\textsuperscript{22} that acts as a specific P-selectin inhibitor. Appropriate amounts of this antiselectin agent were infused over the first 5 minutes of reperfusion. Finally, we perfused 5 rat hearts with 100×10\textsuperscript{6} PMNs alone isolated from blood, as described above.

**Determination of Cardiac Tissue Myeloperoxidase**

Myocardial tissue myeloperoxidase (MPO), an enzyme that occurs virtually exclusively in neutrophils, was determined as described previously.\textsuperscript{23} One unit of MPO is defined as that quantity of enzyme that hydrolyzes 1 mmol peroxide per minute at 25°C. The assays were performed without knowledge of the group in which each sample originated.

**Histology and Immunohistochemistry**

In addition to the hearts used for assessment of left ventricular function, other rat hearts were perfused to determine the number of PMNs infiltrating into the heart by histological methods. After 45 minutes of reperfusion, hearts were removed from the perfusion apparatus and placed in 4% paraformaldehyde overnight at 4°C. The heart was cut into sections and dehydrated in graded acetone washes at 4°C. Tissue sections were embedded in plastic (Immunobed; Polysciences Inc), and 4-μm-thick sections were cut and transferred to Vectabond-coated slides (Vector Laboratories, Inc). The slides were soaked in 95% ethanol for 10 minutes to remove some of the plastic embedding material and to allow staining of the tissue. After the 10-minute ethanol wash, the tissue sections were stained with either hematoxylin in solution, Gill No. 3 (Sigma) for 10 minutes or Giemsa stain (Sigma) for 3 minutes. The slides were then observed microscopically, and the numbers of PMNs and platelets were counted and tallied. Five fields from each of 2 slides were counted from each heart, and 3 rats were studied in each group.

Immunohistochemistry for P-selectin was performed on tissue sections according to previously described techniques in 3 to 4 hearts from each group.\textsuperscript{11} The basic method used was the avidin-biotin immunoperoxidase technique with monoclonal antibody PB1.3 as the monoclonal antibody directed against P-selectin. Positive staining was defined as a coronary microvessel displaying brown reaction product on >50% of the circumference of its endothelium. Fifty vessels per tissue sample were examined in each of 3 or 4 hearts per group.
3106 PMNs were coperfused with 75,000 platelets, the final LVDP was 38 ± 6 mm Hg in the presence of 100,000 platelets. Thus, the final LVDP was 3% (ie, not significantly different from initial). This proischemic effect represents a synergistic relationship between platelets and neutrophils, because their effects are 3 times greater than the additive effects of platelets and PMNs given alone (Figure 1). Nevertheless, perfusion under these conditions with a SLεX-OS, a selectin-blocking agent, prevented these effects on coronary flow. Similarly, perfusion with sPSGL-1 at 5 μg/mL resulted in a decrease in coronary flow of only 5 ± 3%, a value not significantly different from that of the SLεX-OS–treated hearts.

Similar results were obtained with regard to left ventricular cardiodynamics (ie, LVDP, Figure 2). LVDP decreased by 10 ± 2% in hearts perfused with PMNs and 11 ± 2% in hearts perfused with platelets (P < 0.05 from initial) (Figure 2). However, perfusion with both PMNs and platelets resulted in a decline in LVDP of 54 ± 3% (P < 0.001 from initial and from all other groups), again indicating a marked degree of synergy between PMNs and platelets. Moreover, addition of SLεX-OS to the perfusate containing both PMNs and platelets completely blocked this cardiac dysfunction exemplified by the decline in LVDP (Figure 2). In 5 additional rat hearts perfused with sPSGL-1 at 5 μg/mL, the decline in LVDP was only 2 ± 2%.

As would be expected, the first derivative of LVDP (ie, dP/dt max) exhibited the same relationships as did the basic LVDP data (Figure 3). Thus, coronary perfusion with PMNs or with platelets resulted in decreases in dP/dt max of 5 ± 3% and 4 ± 3% (ie, not significantly different from initial), whereas the combination of platelets and PMNs resulted in a
reduction in dP/dt\text{max} of 55-4% (P<0.001 from initial or any other group), reinforcing the dramatic degree of synergy between PMNs and platelets in inducing cardiac dysfunction. Perfusion with the antiselectin agent (ie, SLe\textsuperscript{x}-OS) in combination with platelets and PMNs attenuated the decrease in dP/dt\text{max} to 12-5% (P<0.001 from I/R+PMNs+platelet) (Figure 3). In 5 additional rat hearts perfused with sPSGL-1 at 5 μg/mL, the decrease in dP/dt\text{max} was only 6-4%.

We also collected the hearts at the end of the reperfusion period, froze them at -70°C, and analyzed cardiac tissue for MPO activity as an index of accumulated PMNs. No MPO activity could be detected in control hearts or any I/R heart not perfused with PMNs, thus indicating that there are very few resident PMNs in the perfused rat hearts. However, Figure 4 shows that perfusion of I/R hearts with 75×10\textsuperscript{6} PMNs increased MPO activity significantly (P<0.05 from ischemia/reperfusion alone). Furthermore, coperfusion of I/R hearts with PMNs and platelets increased the MPO activity 3-fold (P<0.02 from I/R+PMNs). However, addition of SLe\textsuperscript{x}-OS to the PMNs and platelets perfusing I/R hearts dramatically attenuated MPO activity to values below those observed for I/R+PMNs alone (Figure 4).

As an additional verification of the MPO activity, we performed histological analysis of perfused rat hearts and counted PMNs and platelets in these sections. Figure 5 summarizes these results. Essentially no resident leukocytes were observed in perfused rat hearts subjected to ischemia-reperfusion under the conditions of the current protocol. However, when the hearts were perfused with 75×10\textsuperscript{6} PMNs, >100 PMNs could be detected per field on each section. The percentage of intravascular PMNs was 83%. No extravascular platelets were observed. Perfusion with 75×10\textsuperscript{6} PMNs and 100×10\textsuperscript{6} platelets resulted in significantly greater numbers of both PMNs and platelets on each microscopic field (P<0.001), indicating a synergistic interaction between the 2 blood cell types. Moreover, coinfusion of the SLe\textsuperscript{x}-OS along with the platelets and PMNs essentially abolished accumulation of both cell types (P<0.001 from I/R+PMNs+platelets). Similar results were obtained with the sPSGL-1-perfused hearts. The MPO activity was 0.08±0.01 U/g heart dry weight.

Figure 2. Initial and final LVDP expressed in mm Hg in isolated perfused rat hearts subjected to 20 minutes of global total ischemia and reperfusion. Ischemic hearts were reperfused in the presence (PMNs, 75×10\textsuperscript{6} and platelets, 100×10\textsuperscript{6}) or absence of PMNs and/or platelets (Plats). PMNs markedly decreased recovery of hearts throughout reperfusion, which was facilitated by platelets. All values are expressed as mean±SEM. Numbers of hearts are at bottom of bars. *P<0.05, ***P<0.001.

Figure 3. Initial and final values for first derivative of LVDP (±dP/dt\text{max}) expressed in mm Hg/s in rat hearts subject to ischemia and reperfusion. Ischemic hearts were perfused in the presence or absence of PMNs and/or platelets (Plats). PMNs caused a significant impairment of hearts as seen in depression of ±dP/dt\text{max}, aided by platelets. All values are expressed as mean±SEM. Numbers of hearts are at bottom of bars. ***P<0.001.
wt, a value not significantly different from that obtained with SLeX-OS.

Figure 6 illustrates the degree of expression of P-selectin on coronary venules in perfused rat hearts. No P-selectin expression was observed in any nonischemic perfused rat hearts. Moreover, very low expression was observed in I/R rat hearts perfused without blood cells (ie, 1% of the vessels). The presence of $75 \times 10^6$ PMNs or $50 \times 10^6$ platelets increased P-selectin expression modestly, but perfusion with both PMNs and platelets resulted in a marked increase in P-selectin expression on the coronary vascular endothelium ($P<0.01$). This enhanced expression of P-selectin was markedly attenuated by coperfusion with the SLeX-OS, the antiselectin agent ($P<0.001$). One can therefore conclude that P-selectin upregulation on the I/R coronary vascular endothelium occurs in the presence of platelets and neutrophils and that this is attenuated by SLeX-OS, which inhibits selectin-mediated interaction between platelets and PMNs, platelets and the endothelium, and PMNs and the endothelium.

**Discussion**

The results obtained in this study clearly point toward 2 important conclusions. First, circulating neutrophils at the time of reperfusion contribute significantly to postreperfusion cardiac contractile dysfunction. Second, blood platelets act in a cooperative manner, synergizing with PMNs to exacerbate this cardiac contractile dysfunction. Although we believe that this is the first report of synergism between platelets and neutrophils in promoting postperfusion cardiac dysfunction, other reports suggesting PMN-platelet interaction in patients with coronary artery disease have recently appeared.15, 19 In this regard, leukocyte-platelet binding has recently been shown to occur in patients either with unstable angina19 or after acute myocardial infarction.15 The basis for this cell-to-cell interaction is not known but may involve P-selectin upregulation on platelet or endothelial cell membranes.24 This concept is consistent with the finding that thrombin stimulation of platelets leads to increased expression of proinflammatory cytokines (eg, interleukin-1β and interleukin-8) by

![Figure 4](http://circ.ahajournals.org/)

**Figure 4.** Cardiac MPO activity in cardiac tissue samples obtained from I/R rat hearts, with PMNs and/or platelets (Plats). MPO activity is expressed in U/g wet wt tissue. All values are mean±SEM of 7 hearts. SLeX-OS significantly inhibited increased MPO activity in hearts perfused with platelets and PMNs.

![Figure 5](http://circ.ahajournals.org/)

**Figure 5.** Quantification of histological staining of cells (PMNs and platelets) in isolated perfused rat hearts after ischemia/reperfusion and reperfusion with PMNs and/or platelets (Plats). Platelets markedly increased amount of PMNs in myocardium compared with PMNs alone at same dose. Five fields were counted on 2 slides of each rat of 3 rat hearts used in each group. Perfusion of I/R rat hearts with PMNs+platelets significantly enhanced accumulation of both cell types ($P>0.001$), which was totally abolished by coperfusion with SLeX-OS ($P<0.001$).

![Figure 6](http://circ.ahajournals.org/)

**Figure 6.** P-selectin expression in coronary vascular endothelium of isolated perfused rat hearts. All values are mean±SEM of 50 sections each in 3 to 4 hearts/group. A positive vessel is one in which $>50\%$ of its endothelial surface exhibits a peroxidase brown reaction product. Perfusion with platelets (Plats)+PMNs significantly upregulated coronary endothelial P-selectin expression in I/R hearts ($P<0.001$). This was significantly attenuated by SLeX-OS ($P<0.001$).
Thus, P-selectin may mediate platelet-neutrophil interaction after ischemia-reperfusion, such as that observed in the perfused rat hearts used in the present study. Another possibility is that leukocytes and platelets interact and cooperate metabolically to facilitate transcellular biosynthesis of some mediator of inflammation, as is known to occur in the case of the leukotrienes.26

Our results add to our knowledge of cell-to-cell interaction in the pathophysiology of reperfusion injury in several new ways. First, the interaction between PMNs and platelets is a synergistic one in which the response of the 2 cell types together is greater than that which would be predicted by the same numbers of the 2 cell types perfused separately. This synergism is clearly suggestive of a humoral interaction amplifying the actions of the 2 cell types, an effect that could be mediated, at least in part, by the selectin family of adhesion glycoproteins.

Second, we have provided evidence that blockade of the selectin family can markedly attenuate this synergism between platelets and neutrophils. This was demonstrable with an SLe\(\alpha\)-OS, which blocks one of the major low-affinity ligands for the selectins,26 and by a soluble PSGL-1, which blocks the major high-affinity ligand for P-selectin.27 SLe\(\alpha\)-OS agent has previously been reported to exert beneficial effects in myocardial ischemia-reperfusion in cats and dogs,24,29 even up to 48 hours after reperfusion. These beneficial effects include reduced myocardial necrosis, fewer neutrophils infiltrating the heart, preserved coronary endothelial function,28 better-maintained myocardial contractility,28,29 and enhanced coronary blood flow.28 Similarly, sPSGL-1 has been shown to be effective in attenuating renal tissue injury in a rat model of renal ischemia-reperfusion.22 These antiselectin agents could exert protective effects in one of several ways. Initially, these agents could have inhibited platelet P-selectin-mediated effects between platelets and neutrophils or between platelets and endothelial cells. Alternatively, they could have inhibited endothelial P-selectin-mediated effects between endothelial cells and neutrophils. Probably, the overall protective effect of the antiselectin agents is a combination of all of these possibilities, because all 3 cell types are intimately involved in the pathophysiology of ischemia/reperfusion.20 Clearly, the adherence of PMNs to the endothelium is central to much of the ensuing pathophysiology,7–9 whether it occurs as single PMNs or clusters of platelet-neutrophil complexes.

The third major conclusion derived from these studies is that blood cells and mediators on their surface or released by these cells can induce significant effects on myocardial contractility in sensitized hearts (ie, I/R). Thus, in our model, myocardial ischemia/reperfusion in the absence of blood cells produced no deficit in cardiac contractility. When PMNs alone or platelets alone were added to the system, modest deficits in contractility occurred. PMNs isolated from the blood were comparable to elicited PMNs, indicating that partially activated PMNs contain sufficient mediators to produce significant cardiac dysfunction. Only when both cell types were given together at reperfusion was there a marked decline in coronary flow and cardiac contractility. Moreover, a comparable degree of cooperativity occurred between platelets and PMNs with ratios of 1:3:1 and 10:1. This suggests that the smaller numbers of circulating platelets are sufficient for this cooperativity between cell types. This could not be attributed to the mere presence of PMNs and platelets, because these cells together in nonischemic perfused rat hearts were without any effect on contractility or coronary flow. The dysfunction is indicative of an activation process that is dependent on reperfusion of an ischemic vasculature.

One may speculate as to the cellular mechanism of this PMN-platelet cooperativity in producing postreperfusion cardiac dysfunction. Several possibilities exist. First, PMN or platelet aggregates or PMN-platelet complexes could obstruct flow in significant numbers of coronary microvessels and plug up these vessels, thus interfering with the normal distribution of coronary flow to the cardiac myocytes. With regard to this possibility, we saw no evidence of obstruction in any of the histological sections we observed. However, we did observe small numbers of homotypic (ie, platelet-platelet or PMN-PMN) or heterotypic (ie, PMN-platelet) aggregates, but these were scattered and were not large enough to physically obstruct coronary vessels. There is evidence of this plugging up of vessels in vivo,4 but under the conditions of our experiments, this did not occur. With regard to proinflammatory humoral mediators, both platelets and PMNs are rich in potential humoral candidates. Platelets have abundant prothrombotic and vasoconstrictor agents, including thrombin, histamine, thromboxane A\(_2\), and platelet factor 4. Several of these humoral agents could contribute to the reduced coronary flow observed in the present experiments, which could lead to cardiac contractile dysfunction. Many of these agents also upregulate P-selectin expression by translocation of P-selectin protein from \(\alpha\)-granules to the surface of the platelet membrane.21,32 Moreover, neutrophils release a variety of cytokines, proteases, lipid mediators, and oxygen-derived free radicals50–52 that can interact with platelet-derived mediators. This combination of mediators could exacerbate both the reduced coronary flow and the cardiac contractile dysfunction observed in the present experiments.

P-selectin expressed on platelet membranes could bind neutrophils to platelets and both cell types to the endothelium, which also expresses P-selectin on its surface translocated there from Weibel-Palade bodies.25 We observed a significant increase in P-selectin expression on I/R coronary vascular endothelium in the presence of PMNs and platelets, and this was markedly attenuated by SLe\(\alpha\)-OS. Under the conditions of the present flow-through experiment (ie, no recirculation), we could not accurately measure release of humoral agents, which could upregulate P-selectin. However, our results are consistent with the presence of humoral mediators. In any case, we have clear data to support the third hypothesis, that platelets and PMNs synergistically promote enhanced endothelial adhesiveness and eventful transendothelial migration of PMNs into the I/R myocardium. We observed both significantly elevated cardiac MPO activity, a specific marker of neutrophils, and increased numbers of PMNs microscopically in ischemic hearts reperfused with both cell types compared with those reperfused with either cell type alone. The net result is to augment and exacerbate the effects of neutrophils on propagating cardiomyocyte injury after reperfusion of the...
ischemic myocardium. One factor contributing to this phenomenon could be the ability of activated PMNs to stimulate platelet function. Therefore, these results suggest that either antiplatelet or antineutrophil agents may provide some benefit against reperfusion injury. Moreover, dual antiplatelet and antineutrophil therapy or an agent that addresses both cell types may be of particular utility in myocardial ischemia-reperfusion.

These findings are of potential clinical relevance but clearly must be confirmed in an in vivo model of reperfusion injury before further consideration of their implications can be addressed. Nevertheless, there are suggestions that these findings will be shown to be clinically relevant, as has been suggested in transplant rejection and stroke.

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References
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