Myocardial infarction (MI) represents the most frequent cardiovascular event in the western world and is responsible for a large part of cardiovascular deaths. Currently, improved clinical management of acute MI has significantly lowered immediate mortality. However, surviving patients are facing another major complication, the development of heart failure, in part as a consequence of inadequate infarct healing.

**An Adequate Healing Process Is Critical**

This repair process is of crucial importance to promote the healing of damaged myocardial tissue by removal of dead cells, replacement of necrotic areas by connective tissue, and remodeling of myocardial structure and geometry to ultimately compensate for the loss of cardiomyocytes (1). Several phases of infarct healing have been described that include an initial inflammatory response, followed by the influx of neutrophils and monocyte/macrophages, which orchestrate wound-healing leading to the formation of a cardiac scar within approximately 3 weeks. Although these compensatory or regulatory mechanisms are beneficial at first, their prolonged persistence can be deleterious as well (1). Therefore, as more patients survive the acute phase of MI and enter the period of infarct healing, a better understanding of the mechanisms that promote “optimal” infarct healing is needed.

**Role of Macrophages in Healing**

Monocyte-derived macrophages are thought to take a central role in the regulation of infarct healing. They facilitate the wound-healing process (2)—by removing necrotic cardiac myocytes and apoptotic neutrophils as well as in the secretion of cytokines, chemokines, and proteases. In fact, a recent study showed that macrophage depletion with clodronate-containing liposomes impaired wound-healing in a murine MI model (3), and injection of activated macrophages into the ischemic myocardium has been found to improve myocardial healing and preserve left ventricular (LV) function in mice (4). Thus, a macrophage-mediated inflammatory response is critical for an adequate tissue repair. By contrast, if not properly orchestrated, it might also have a detrimental effect. Indeed, leukocyte counts in the blood of MI patients have been shown to predict outcome (5–7), and patients with monocytosis were found to display a higher risk of developing LV dilation, resulting in an impaired ejection fraction (6,7). However, these studies do not address the heterogeneous nature of monocyte/macrophages and their differences that might explain their dual role in myocardial healing.

**Divergent Functions of Monocyte Subsets**

Recently, the heterogeneity of circulating monocytes has become the focus of attention in many pathological systems. In the peripheral blood of mice, 2 major monocyte subsets have been described and distinguished according to their expression of cell surface proteins (8). The so-called “inflammatory” monocytes express high levels of Ly-6C (Ly-6C\textsuperscript{hi}), the monocyte chemoattractant protein (MCP)-1 receptor CCR2, the adhesion molecules L-selectin (CD62L), alpha-M integrin beta 2 (CD11b), and low levels of the fractalkine receptor CX3CR1. They migrate to inflamed tissues, principally in response to the CCR2 ligand MCP-1. The second subset of monocytes has been termed “resident” monocytes, because they are typically found in both resting and inflamed tissues. Although their functions still remain unclear, they have recently been suggested to play an important homeostatic role in patrolling healthy tissues through long-range crawling on the resting endothelium in the steady state. These monocytes are defined by the high expression of CX3CR1 and alpha-L integrin beta 2 (CD11a) and lack of expression of Ly-6C (Ly-6C\textsuperscript{lo}), CCR2, and CD62L (8).

**Monocyte Subsets in Infarct Healing**

In a preceding study using a coronary ligation model to induce MIs in mice, Nahrendorf et al. (9) found that these 2 monocyte subsets are sequentially recruited into the infarcted area in a process that is orchestrated by the differential expression of the chemokines MCP-1 and fractalkine, respectively. Thus, MI repair is characterized by a biphasic response, in which phase I is dominated by Ly-6C\textsuperscript{hi} monocytes that exhibit phagocytic, proteolytic, and inflammatory properties, whereas phase II is dominated by the anti-inflammatory Ly-6C\textsuperscript{lo} monocytes that express high levels of vascular endothelial growth factor. Therefore, the 2 monocyte subsets were hypothesized to have divergent roles...
during the wound-healing process. The Ly-6Chi monocytes digest damaged tissue, whereas Ly-6Clo monocytes promote healing via myofibroblast accumulation, collagen deposition, and promotion of angiogenesis (Fig. 1).

Importantly, in humans different monocyte subsets have also been indentified on the basis of the expression of CD14 and CD16 (8). The CD14⁺/H11001 CD16⁺/H11002 monocytes represent the major subset in the peripheral blood and express CCR2 but only low levels of CX3CR1, resembling the “inflammatory” Ly-6Chi monocytes in mice. They are potent phagocytes and produce large amounts of proinflammatory cytokines. In contrast, a smaller subset of monocytes is CD14⁻/H11001 CD16⁻/H11001 and exclusively express CX3CR1, sharing a phenotype similar to Ly-6Clo monocytes in mice. Recently, Tsujioka et al. (10) studied the dynamics of these monocyte populations in humans with acute MI and—in analogy to the murine data previously discussed—found an initial rise of “inflammatory” CD14⁺/H11001 CD16⁻/H11002 monocytes followed by a sequent mobilization of circulating monocytes with the CD14⁻/H11001 CD16⁻/H11001 phenotype. In fact, peak levels of the inflammatory subset were found to be negatively correlated with myocardial salvage after 7 days and recovery of LV function after 6 months.

Monocytosis Impairs Infarct Healing in Mice

Nahrendorf et al. also studied infarct healing in cholesterol-fed apolipoprotein E knockout (apoE⁻/⁻) mice, which are known to have increased levels of circulating Ly-6Chi monocytes (11). The apoE⁻/⁻ mice were found to have increased signs of impaired infarct healing, suggesting a central role for monocytes in the outcome of MI repair. However, it was not clear whether this impairment was due to the monocytosis itself, a disturbed transition from phase I to phase II, an altered monocyte function in general, or other factors related to the apoE deficiency.

Now, Panizzi et al. (12) shed light on these important questions. With their established MI model, the authors found >10× higher levels of Ly-6Chi monocytes and a significantly increased ratio of Ly-6Chi/Ly6Clo monocytes in the infarct areas of apoE⁻/⁻ mice during phase II (day 5 after MI), which is usually dominated by Ly-6Clo monocytes. They could further document, consistent with the increased levels of Ly-6Chi monocytes, increased proteolytic and phagocytic activity in the infarcted tissues of apoE⁻/⁻ mice compared with nonatherosclerotic mice, despite equal infarct size. Consequently, LV remodeling was impaired in apoE⁻/⁻ mice, resulting in an increased end-diastolic volume and significantly worsened ejection fraction. Importantly, with in vivo administered fluorescent probes for proteolytic and phagocytic activity followed by flow-cytometric analyses of cells of the digested infarct tissue, the majority of the proteolytic activity could be attributed to the Ly-6Chi monocytes. The contribution of neutrophils, which also possess high proteolytic activity and are equally increased in atherosclerotic mice, could be ruled out by depletion with a specific antibody, which did not rescue this phenotype. Finally, to demonstrate that the persistent presence of Ly-6Chi monocytes with proteolytic activity, as found in atherosclerotic mice, is indeed responsible for deregulated wound-healing, the authors employed a model of lipopolysaccharide-induced monocytosis in wild-type mice. In this model, the isolated increase of Ly-6Chi monocytes in the blood and the infarcted tissues resulted in an analogous phenotype as seen in apoE⁻/⁻ mice. Thus, the presence of increased numbers of inflammatory monocytes in the infarct could be directly linked to the outcome of LV remodeling.

Figure 1 Finding the Right Balance: Distinct Monocyte Subsets Mediate Resolution and Repair in Myocardial Infarction

CCR2 = chemokine (C-C motif) receptor 2; CX3CR1 = chemokine (C-X3-C motif) receptor 1; MCP = monocyte chemoattractant protein; PMN = neutrophils.
These data identify Ly-6Chi monocytes as the culprit cells for impaired infarct healing, although the exact mechanism for their persistence in infarcted areas of atherosclerotic mice remains to be shown. It is still unclear whether their dominance throughout the healing process simply reflects the increased circulating levels seen in these mice or whether it is a consequence of increased and persistent local expression of specific ligands for CCR2, most prominently MCP-1. Moreover, it is not clear whether the dynamic expression of chemokines that have been shown to be crucial for the recruitment of both monocyte subsets and the orchestration of a proper healing process is altered in atherosclerotic mice. What are the factors that control this biphasic chemokine expression during the healing process? How is phase I ended? And what initiates phase II of infarct healing? It might well be that the ongoing chronic inflammation in atherosclerotic mice generates an environment that significantly hampers this transition, irrespective of the high levels of circulating monocytes. To answer this, a more detailed understanding of the physiological healing processes that occur after an MI and the factors modulating it is needed.

Such knowledge will likely offer potential therapeutic targets to modulate or correct a deregulated wound-healing process. Of interest, statin treatment of atherosclerotic apoE<sup>−/−</sup> mice has been shown to reduce Ly-6C<sup>hi</sup> monocyte counts from peripheral blood and bone marrow (11) and to down-regulate CCR2 expression of circulating monocytes in humans (13). Nevertheless, the biphasic response of proper infarct healing and the necessity for the sequential recruitment of both monocyte subsets requires an exact timing of any given intervention and will likely be the most challenging part for translating these findings to humans.

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