The Role of Cardiac Fibroblasts in the Transition from Inflammation to Fibrosis Following Myocardial Infarction

Frans A. van Nieuwenhoven[1] and Neil A. Turner[2]*

[1]Department of Physiology, Cardiovascular Research Institute Maastricht (CARIM), Maastricht University, Maastricht, The Netherlands.

[2]Division of Cardiovascular Medicine, and Multidisciplinary Cardiovascular Research Centre (MCRC), University of Leeds, Leeds, UK.

*Corresponding author: Dr Neil A. Turner, Division of Cardiovascular Medicine, Worsley Building, Clarendon Way, University of Leeds, Leeds LS2 9JT, UK.

Tel: +44(0)113-3435890. Fax: +44(0)113-3434803. E-mail: n.a.turner@leeds.ac.uk

ABSTRACT

Cardiac fibroblasts (CF) play a pivotal role in the repair and remodeling of the heart that occurs following myocardial infarction (MI). The transition through the inflammatory, granulation and maturation phases of infarct healing is driven by cellular responses to local levels of cytokines, chemokines and growth factors that fluctuate in a temporal and spatial manner. In the acute inflammatory phase early after MI, CF contribute to the inflammatory milieu through increased secretion of proinflammatory cytokines and chemokines, and they promote extracellular matrix (ECM) degradation by increasing matrix metalloproteinase (MMP) expression and activity. In the granulation phase, CF migrate into the infarct zone, proliferate and produce MMPs and pro-angiogenic molecules to facilitate revascularization. Fibroblasts also undergo a phenotypic change to become myofibroblasts. In the maturation phase, inflammation is reduced by anti-inflammatory cytokines, and increased levels of profibrotic stimuli induce myofibroblasts to synthesize new ECM to form a scar. The scar is contracted through the mechanical force generated by myofibroblasts, preventing cardiac dilation. In this review we discuss the transition from myocardial inflammation to fibrosis with particular focus on how CF respond to alterations in proinflammatory and profibrotic signals. By furthering our understanding of these events, it is hoped that new therapeutic interventions will be developed that selectively reduce adverse myocardial remodeling post-MI, whilst sparing essential repair mechanisms.

Key words: cardiac fibroblasts; heart; inflammation; fibrosis; myocardial remodeling

1. Introduction

Myocardial infarction (MI)[1] results from ischaemia of an area of myocardial tissue subserved by an occluded coronary artery. The consequent injury and death of cardiomyocytes triggers an acute inflammatory response that initiates the repair of the myocardium through a series of well orchestrated events (summarized in Fig. 1 and Fig. 2). The inflammatory phase is associated with rapid local increases in proinflammatory cytokines and chemokines, such as interleukin (IL)-1, IL-6, IL-8 and tumor necrosis factor α (TNFα), and neutrophil infiltration (Frangogiannis et al., 2002; Frangogiannis and Entman, 2005). Cardiac fibroblasts (CF) are the most prevalent resident myocardial cell type, accounting for up to two-third of heart cells (Porter and Turner, 2009). Although traditionally known for being essential regulators of extracellular matrix (ECM) remodeling, it is becoming increasingly apparent that CF play a central role in many aspects of post-MI remodeling of the heart (Porter and Turner, 2009). Recent evidence suggests that CF may contribute to the early inflammatory phase of remodeling by secreting several important proinflammatory cytokines and chemokines (Turner et al., 2009; Turner et al., 2010; Turner et al., 2011).

Approximately 3 days after MI, the infarct area is infiltrated by macrophages and phenotypically modified fibroblasts (myofibroblasts), that form granulation tissue in which ECM-degrading proteases of the matrix metalloproteinase (MMP) family and pro-angiogenic molecules such as vascular endothelial growth factor (VEGF) are up-regulated (Jourdan-Lesaux et al., 2010; Lindsey and Zamilpa, 2012). The granulation stage of remodeling is characterized by the clearance of dead cells, the breakdown of ECM and stimulation of angiogenesis to revascularize the ischemic tissue.

Transition from the inflammatory phase to the fibrotic stage of remodeling ("maturation") typically occurs 3-7 days post-MI depending on the species (Dewald et al., 2004) and appears to be mediated by increased production of anti-inflammatory and

profibrotic factors such as transforming growth factor-β (TGFβ) and IL-10, derived from both infiltrating immune cells and myocardial cells. In response to such factors, myofibroblasts synthesize increased amounts of structural ECM proteins, particularly type I and III fibrillar collagens, to facilitate scar formation. This response is essential to replace the areas devoid of cardiomyocytes and prevent rupture of the myocardial wall. Balanced ECM remodeling is crucial, since inadequate strength of the myocardial wall will lead to cardiac dilation, while excessive or prolonged ECM accumulation can result in fibrosis. While cardiac dilation primarily affects systolic function, fibrosis stiffens the myocardium and mainly hampers the diastolic filling phase.

Myofibroblasts express increased levels of contractile proteins such as alpha-smooth muscle actin (αSMA), vimentin and focal adhesion proteins that permit them to contract the scar area to enable effective wound healing (van den Borne et al., 2010). In contrast to wound healing in other tissues and organs (e.g. skin) in which the myofibroblasts are effectively removed from scar tissue by apoptosis, cardiac myofibroblasts appear to remain within the healed infarct area for many years (Palatinus et al., 2010; van den Borne et al., 2010). Whether myofibroblast persistence is beneficial or detrimental to the integrity of the healed infarct remains to be firmly established.

Effective infarct healing involves the coming together of both resident and infiltrating cell types in a highly spatially and temporally coordinated manner. Given that infarct size is a major predictor of post-MI mortality (Burns et al., 2002), the transition from the early inflammatory phase of remodeling (which increases infarct area) to the fibrotic maturation phase of remodeling (which limits infarct expansion) is paramount for effective healing and reduced secondary complications. This review focuses on the key role played by CF in this transition.

2. Cardiac fibroblasts

Under normal physiological conditions, CF are relatively inert cells that maintain myocardial ECM homeostasis. However, in response to cardiac injury or stress, CF can adopt a specialized myofibroblast phenotype (van den Borne et al., 2010) that enables the cells to respond more effectively to environmental stimuli. This phenotypic plasticity of fibroblasts in the heart allows them to migrate into the infarcted area in response to chemotactic stimuli, proliferate in response to mitogenic factors, and differentiate into myofibroblasts that drive aggressive remodeling of the ECM and wound contraction, enabling rapid and effective repair of the cardiac interstitium (see Fig. 2).

Very recently, reprogramming of cardiac fibroblasts into functional cardiomyocytes using specific transcription factors or micro-RNAs was reported (Ieda et al., 2010; Qian et al., 2012; Song et al., 2012; Jayawardena et al., 2012). This revolutionary technique holds great promise for replacing necrotic cardiomyocytes following MI by generating functional cardiac muscle tissue from resident CF. If applicable after human MI, this approach could reduce infarct size and adverse remodeling and improve cardiac functional recovery.

2.1. Myofibroblast phenotype

It has long been accepted that cardiac myofibroblasts can be derived from mature resident CF, but there is now increasing evidence that they can also originate from endothelial cells, epithelial cells, pericytes, circulating bone marrow-derived progenitor cells (fibrocytes) and mesenchymal stem cells (Carlson et al., 2011; Zeisberg et al., 2007; Keeley et al., 2010; Haudek et al., 2006). Irrespective of their origin, myofibroblasts are characterised by increased expression of specific contractile proteins (αSMA, vimentin), focal adhesion proteins (paxillin, tensin, integrins), cell surface receptors (TβRII, AT1R, Frizzled) and ECM proteins (collagen I, collagen III and the ED-A splice variant of fibronectin) (van den Borne

et al., 2010; Tomasek et al., 2002; Daskalopoulos et al., 2012). Many of these molecular changes confer increased tensile characteristics on the myofibroblast cell, allowing it to efficiently contract the surrounding ECM and facilitate effective post-MI scar formation and thinning (van den Borne et al., 2010; Tomasek et al., 2002).

The profibrotic cytokine TGFβ is considered to be the prime inducer of the mature myofibroblast phenotype (Dobaczewski et al., 2011; Tomasek et al., 2002). Other factors that contribute to myofibroblast differentiation include mechanical tension and ED-A fibronectin (Tomasek et al., 2002) and more recently, roles for Type VI collagen (Naugle et al., 2006) and Wnt/frizzled signaling (Laeremans et al., 2010) have been described in CF. Although not particularly well studied in the heart, there is evidence that TGFβ-induced differentiation of myofibroblasts from non-cardiac sources can be opposed by the action of proinflammatory cytokines, including IL-1β and TNFα (Shephard et al., 2004; Liu et al., 2009).

Interestingly, when grown *in vitro* in rigid culture vessels, CF spontaneously undergo transition to a myofibroblast phenotype which is maintained or enhanced with passaging (Mughal et al., 2009; Santiago et al., 2010; Teunissen et al., 2007). This *in vitro* phenotype shares many similarities with that of myofibroblasts found in the infarct area *in vivo* (Santiago et al., 2010). In addition to enhanced contractile, focal adhesion and receptor proteins, cultured myofibroblasts are less motile than their fibroblast counterparts, reflecting a change from migratory to synthetic and contractile function (Santiago et al., 2010).

2.2. Regulation of extracellular matrix turnover by cardiac fibroblasts

CF regulate myocardial ECM turnover by balancing the synthesis and degradation of ECM components in response to physiological and pathophysiological stimuli. As the key source of ECM molecules in the heart, (myo)fibroblasts can synthesize a wide range of structural matrix proteins (e.g. fibrillar collagens, laminins, fibronectin), as well as matricellular

proteins; a group of ECM-regulatory proteins that includes connective tissue growth factor (CTGF/CCN2), thrombospondins, tenascins, osteopontin/SPP1 and osteonectin/SPARC (Frangogiannis, 2012). These matricellular proteins are extracellular proteins that do not serve structural roles, but rather modulate cell-matrix interaction and cellular function (Bornstein and Sage, 2002). For example, CCN2 can bind TGFβ1 and enhance its signaling activity (Abreu et al., 2002).

Conversely, CF are also a rich source of proteases, including the MMP family of enzymes that together are capable of degrading all the protein components of the myocardial ECM (Spinale, 2007). CF express several different MMPs, including MMP-1, -2, -3, -9 and -14, and their expression and activation can be differentially modulated by various proinflammatory (e.g. IL-1, TNFα) and profibrotic (e.g. TGF-β, Ang II) stimuli (Turner and Porter, 2012). CF can further control the proteolytic activity of MMPs through regulating expression and secretion of TIMPs, the endogenous inhibitors of MMPs, with TIMP-1 and TIMP-2 being the major forms expressed by CF (Brown et al., 2007; Turner et al., 2010). Hence, by coordinating synthesis and activation of structural ECM proteins, matricellular proteins, MMPs and TIMPs, CF are able to precisely control myocardial ECM turnover, and help to compensate for impaired cardiac function following myocardial injury or stress.

2.3. Cardiac fibroblasts as producers of autocrine/paracrine factors

CF represent a rich local source of secreted bioactive molecules that can regulate the function of adjacent cells via autocrine/paracrine signaling networks. These factors include proinflammatory cytokines (e.g. TNFα, IL-1β and IL-6), chemokines (e.g. CXCL1/GROα and CXCL8/IL-8), profibrotic factors (e.g. TGFβ, Ang II and endothelin-1) and molecules regulating neovascularization (e.g. VEGF and ADAMTS1) (Porter and Turner, 2009; Turner et al., 2009; Turner et al., 2010; Turner et al., 2011). By regulating the local concentrations of

such factors, CF are able to modulate not only their own function but that of other cell types (e.g. cardiomyocytes, inflammatory cells, endothelial cells) to coordinate the myocardial remodeling process.

3. Cardiac fibroblast responses to proinflammatory and profibrotic stimuli

3.1. Effect of proinflammatory stimuli on CF

The levels of proinflammatory cytokines TNFα and IL-1 increase rapidly in the infarcted myocardium (Frangogiannis et al., 2002; Ono et al., 1998). TNFα may derive from mast cells (Gordon and Galli, 1990) or fibroblasts (Turner et al., 2009), while IL-1 could initially be released from necrotic cardiomyocytes (Chen et al., 2007).

The IL-1 family of cytokines includes IL-1α and IL-1β which have similar structure, bind to the same membrane receptors and apparently have indistinguishable biological activities (Bujak and Frangogiannis, 2009). In isolated human CF, IL-1α strongly stimulated the expression of the pro-inflammatory cytokines IL-1β, TNFα and IL-6, implicating CF as being important in mediating the post-MI inflammatory response (Turner et al., 2009). Furthermore, IL-1α enhanced the expression of several CXC chemokines and adhesion molecules important in neutrophil infiltration (Turner et al., 2011). A similar stimulatory effect on CF chemokine expression was also described for Oncostatin M, a member of the IL-6 family of cytokines (Lafontant et al., 2006). Moreover, IL-1α induced MMP-1, MMP-3, MMP-9 and MMP-10, indicating that increased levels of this cytokine have important consequences for ECM degradation (Turner et al., 2010). Increased MMP expression and activity was also observed in rat CF stimulated by IL-1β (Brown et al., 2007) or TNFα (Siwik et al., 2000). IL-1 and TNFα reduced total collagen synthesis ([^{3}H]-proline incorporation) in neonatal and adult rat CF suggesting these cytokines may contribute to ventricular dilation (Siwik et al., 2000; Xiao et al., 2008). However, we reported recently that

IL-1 had no direct effect on basal Type I collagen (COL1A1) mRNA expression in human CF measured after 6 h (Turner et al., 2010). In contrast, IL-1 markedly reduced CCN2 mRNA expression at this early time point (Turner, 2011, unpublished data). It is feasible therefore that the longer-term reductions in collagen expression that were previously reported in response to IL-1-stimulated rat CF (Siwik et al., 2000; Xiao et al., 2008) are due to an indirect autocrine/paracrine mechanism involving reduced CCN2 expression.

IL-1 also acts on CF to modulate factors that are important for neovascularization of the ischemic area. Using a focused microarray approach, we identified ADAMTS1 as a gene whose expression was reduced in response to IL-1 stimulation in human CF (Turner et al., 2010). ADAMTS1 is a secreted metalloproteinase that inhibits neovascularization through inhibition of VEGF signaling. Thus, by reducing ADAMTS1 expression and increasing VEGF expression at the level of the CF (Turner et al., 2010), IL-1 may promote angiogenesis in the infarcted myocardium.

Aside from the gene expression responses, IL-1β was shown to inhibit CF proliferation (Palmer et al., 1995) and enhance migration of these cells (Mitchell et al., 2007). By contrast, IL-33, another member of the IL-1 family, inhibited rat CF migration (Zhu and Carver, 2012). Surprisingly, in both studies similar signaling pathways (MAP kinase) were activated (Mitchell et al., 2007; Zhu and Carver, 2012). A further member of the IL-1 family, IL-18, was also shown to moderately increase CF migration and proliferation (Fix et al., 2011). Very recently, IL-17A, belonging to a new family of cytokines, was reported to stimulate CF proliferation and migration (Valente et al., 2012). The net effect of the combination of these cytokines on CF proliferation and migration *in vivo* will be dependent on their relative abundance and activity in the remodeling myocardium.

Taken together, the above reports indicate that CF are actively involved in the inflammatory phase following MI by producing cytokines, chemokines and adhesion

molecules, and contributing to infiltration of immune cells. In addition, under the influence of IL-1-family inflammatory cytokines CF show increased MMP activity, which favors ECM degradation, remodeling and neovascularization. See Fig. 2 for summary.

3.2. Effect of profibrotic stimuli on CF

Various humoral factors including TGFβ, Ang II, CCN2/CTGF, basic fibroblast growth factor (bFGF), platelet-derived growth factor (PDGF), insulin-like growth factor-1 and catecholamines are important in cardiac fibrosis (Leask, 2010; Manabe et al., 2002; Nicoletti and Michel, 1999; Daniels et al., 2009). The best known and most studied pro-fibrotic stimulus in the heart is TGFβ. After MI, TGFβ is strongly induced, and it plays a central role in infarct healing and myocardial remodeling (Bujak and Frangogiannis, 2007; Leask, 2007; Lijnen et al., 2000). Mammals express three structurally similar isoforms of TGFβ (TGFβ1, TGFβ2 and TGFβ3). Although these isoforms are encoded by separate genes, and are thus expressed independently, they signal through the same cell surface receptors, and have identical downstream signaling effects. TGFβ1 is increased rapidly after MI and is highly expressed in the first week, while TGFβ3 shows a delayed induction and is strongly increased after 1 week (Dewald et al., 2004). TGFβ1 expression was found to remain elevated 4 weeks after MI (Chuva de Sousa Lopes SM et al., 2004; Sun et al., 1998).

Initially, TGFβ may play a role in suppressing cytokine expression in the infarct region, thereby inhibiting the inflammatory reaction. In the later phase, TGFβ is critical in differentiation of CF to myofibroblasts and *de novo* synthesis of ECM molecules essential for infarct healing (Bujak and Frangogiannis, 2007). The importance of TGFβ function in infarct healing was elegantly shown by TGFβ inhibition studies (Frantz et al., 2008). Anti-TGFβ-antibodies administered after MI led to further left ventricular dilation and increased mortality (Frantz et al., 2008). Similar effects were reported for suppression of TGFβ in

pressure-overload hearts (Lucas et al., 2010), stressing the importance of TGFβ in promoting collagen deposition. As noted earlier, balanced ECM production and degradation is essential, and inappropriate accumulation of collagen leads to cardiac fibrosis. TGFβ has been shown to be critical in cardiac fibrosis in various experimental models (Lijnen et al., 2000), and inhibition of TGFβ in such models prevents fibrosis (Teekakirikul et al., 2010). Myocardial overexpression of TGFβ1 induces atrial, but not ventricular fibrosis (Nakajima et al., 2000; Verheule et al., 2004). Possibly, this remarkable difference in fibrotic response between atria and ventricles is caused by the abundance of CCN2 in atria, which may enhance the effects of TGFβ (Daniels et al., 2009).

After MI, CF differentiate to myofibroblasts which exhibit increased fibrillar collagen synthesis, αSMA expression and capacity to contract their surrounding ECM. Both Ang II and TGFβ1 induce fibroblast-to-myofibroblast differentiation (Swaney et al., 2005), and are potent stimulators of collagen production (Butt et al., 1995; Lijnen et al., 2000; Chua et al., 1991; Eghbali et al., 1991; Swaney et al., 2005). The effects of Ang II and TGFβ may be related as Ang II can induce TGFβ, and both are involved in an integrated signaling network promoting cardiac remodeling (Leask, 2010; Rosenkranz, 2004). The contraction of ECM by cardiac myofibroblasts is an important process in wound healing, and can be studied *in vitro* by culturing cells in 3D collagen gels. Both Ang II and TGFβ enhance *in vitro* collagen gel contraction by CF (Burgess et al., 1994; Drobic et al., 2007; Lijnen et al., 2003).

Collectively, these studies on the effects of profibrotic stimuli on CF provide strong evidence for differentiation toward myofibroblasts, increased collagen production and enhanced contractile behavior. TGFβ may be the most important driving factor for myofibroblast differentiation and ECM synthesis, but interaction and synergy with other factors (e.g. CCN2) may be essential in infarct healing and in post-MI fibrosis. See Fig. 2 for summary.

3.3. Interaction between proinflammatory and profibrotic stimuli

During the MI healing process, CF are exposed to various autocrine and paracrine signals, the strength and composition of which changes with time after the initial insult. As outlined above, initially proinflammatory signaling prevails. In the granulation and maturation phase of infarct healing, anti-inflammatory/profibrotic factors become more important and lead to suppression of inflammatory mediators and ECM remodeling toward a matured scar tissue. The mechanisms of interaction between the various factors in the different phases of infarct healing are poorly understood, but it is apparent that some of the factors have opposing effects on CF biology (Siwik and Colucci, 2004). For example, IL-1β and TNFα inhibited collagen synthesis in CF, and thus opposed the action of TGFβ (Siwik et al., 2000). Moreover, IL-1 or TNFα inhibited basal or TGFβ-induced expression of the profibrotic matricellular protein CCN2 in human (Abraham et al., 2000; Nowinski et al., 2002) or mouse (Yu et al., 2009) fibroblasts of non-cardiac origin, results that we have since confirmed with IL-1 in human CF (Turner, unpublished data 2011).

Surprisingly few studies have been performed to unravel the effect of combinations of cytokines and growth factors on CF function. The net effect of IL-1β and TGFβ1 on adult rat CF function was studied by Brown and colleagues, who demonstrated that TGFβ1 could inhibit IL-1β-induced MMP-2, -3 and -9 activity and cell migration (Brown et al., 2007). Importantly, TGFβ on its own had no effect on these parameters. TGFβ1 was also reported to prevent CF proliferation in response to the IL-6 family cytokine cardiotrophin-1 (CT-1) (Drobic et al., 2007). TGFβ1 and CT-1 had opposing effects when tested individually, but in combination TGFβ1-mediated effects superseded those of CT-1. TGFβ-induced collagen gel contraction was also inhibited by CT-1 (Drobic et al., 2007). Further evidence for the counterbalancing effects of proinflammatory cytokines and profibrotic cytokines comes from

studies using fibroblasts of non-cardiac origin. For example, IL-1-induced apoptosis in corneal myofibroblasts was prevented by TGFβ1 (Kaur et al., 2009). In a co-culture of keratinocytes and dermal fibroblasts, TGFβ-mediated αSMA induction was suppressed by IL-1α (Shephard et al., 2004). Moreover, blocking the IL-1 receptor potentiated αSMA expression in the fibroblasts, indicating an inhibitory effect of IL-1 on TGFβ-induced myofibroblast differentiation (Shephard et al., 2004). If extrapolation of these data to the MI area is permitted, this would imply that during the early phase following ischemia when pro-inflammatory cytokines are abundant, cardiac myofibroblast differentiation and ECM deposition is hampered (summarized in Fig. 2).

Aside from opposing effects of proinflammatory and profibrotic factors, synergistic upregulation of MMP-9 by IL-1β and either PDGF or bFGF was reported in dermal fibroblasts (Bond et al., 1998). If such synergy also exists in the myocardial infarct, this might have an important role in ECM degradation during the infarct remodeling.

The studies on the effect of combinations of pro-inflammatory and pro-fibrotic factors in CF are still very limited in number and suggest that the relative abundance, activity, balance and interactions between these factors are pivotal in determining the outcome of the different phases of wound healing following MI.

4. Conclusions

The remodeling events that occur following myocardial damage are clearly complex and regulated by temporal and spatial fluctuations in key bioactive molecules, many of which influence CF function or are derived from the CF themselves. *In vitro* cell culture studies have proved useful in characterizing CF responses to individual cytokines and growth factors, and limited evidence is available on how certain combinations of molecules act to modulate CF function. However, more studies are clearly warranted to more accurately define the net

functional effects of multiple stimuli at the level of the CF, in order to more precisely model the *in vivo* scenario. By understanding the interplay between inflammatory and fibrotic signals at the level of individual myocardial cell types, we hope to develop effective therapies that protect essential repair processes post-MI, whilst reducing pathological remodeling.

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FIGURE LEGENDS

Fig. 1. The main phases of post-MI myocardial remodeling. Abbreviations: CF, cardiac fibroblast; CMF, cardiac myofibroblast; ECM, extracellular matrix; GF, growth factor; MMP, matrix metalloproteinase.

Fig. 2. The central role of cardiac fibroblasts in responding to proinflammatory and profibrotic signals. See main text for details. Myocardial infarction causes damage and death of myocytes with subsequent production of damage-associated molecular patterns (DAMPs); molecules that trigger an acute inflammatory response. CF respond by undergoing migration, proliferation and increased expression of proinflammatory cytokines, chemokines and MMPs. Subsequently in the granulation phase, profibrotic stimuli (e.g. TGFβ, PDGF etc) induce CF to differentiate to a myofibroblast phenotype with increased αSMA expression, increased expression of pro-fibrotic factors (e.g. CCN2/CTGF) and enhanced collagen secretion. In the maturation phase, myofibroblasts continue to synthesize ECM proteins and contract the wound area. The relative amounts of proinflammatory and profibrotic stimuli fluctuate at different stages of the post-MI remodeling process, and there is increasing evidence that these two types of stimuli have opposing effects on CF function.

Figure 1

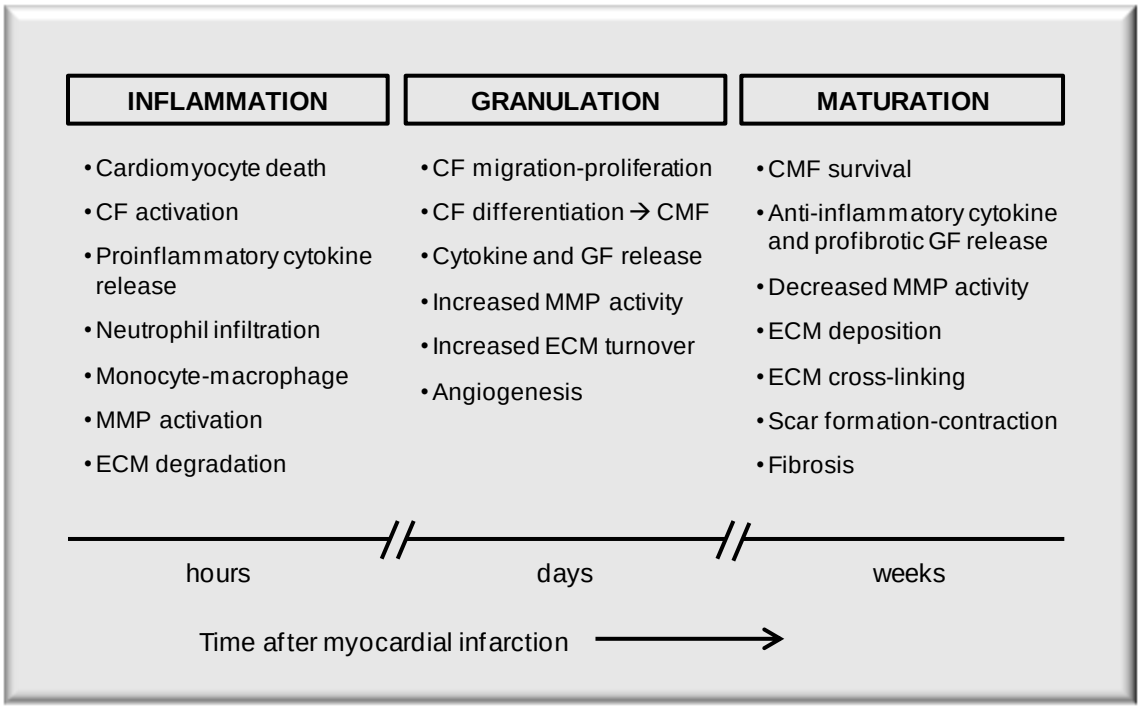
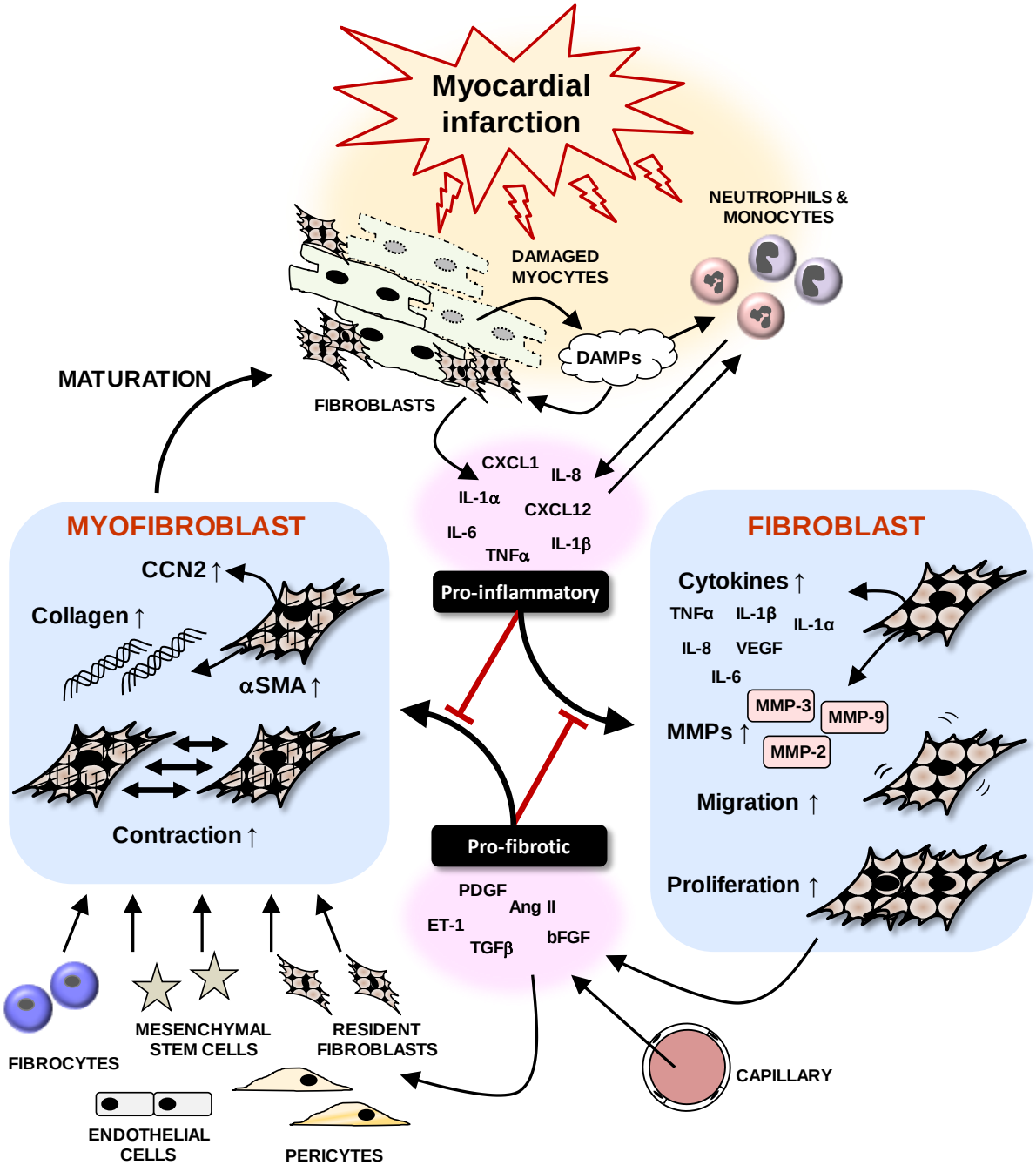

INFLAMMATION
• Cardiomyocyte death
• CF activation
• Proinflammatory cytokine release
• Neutrophil infiltration
• Monocyte-macrophage
• MMP activation
• ECM degradation
GRANULATION
• CF migration-proliferation
• CF differentiation → CMF
• Cytokine and GF release
• Increased MMP activity
• Increased ECM turnover
• Angiogenesis
MATURATION
• CMF survival
• Anti-inflammatory cytokine and profibrotic GF release
• Decreased MMP activity
• ECM deposition
• ECM cross-linking
• Scar formation-contraction
• Fibrosis
hours
days
weeks
Time after myocardial infarction

Figure 2


Myocardial infarction
NEUTROPHILS & MONOCYTES
DAMAGED MYOCYTES
DAMPs
MATURATION
FIBROBLASTS
CXCL1
IL-8
IL-1α
CXCL12
IL-6
TNFα
IL-1β
Pro-inflammatory
MYOFIBROBLAST
CCN2 ↑
Collagen ↑
αSMA ↑
Contraction ↑
FIBROBLAST
Cytokines ↑
TNFα
IL-1β
IL-1α
IL-8
VEGF
IL-6
MMPs ↑
MMP-3
MMP-9
MMP-2
Migration ↑
Proliferation ↑
Pro-fibrotic
PDGF
Ang II
ET-1
TGFβ
bFGF
FIBROCYTES
MESENCHYMAL STEM CELLS
RESIDENT FIBROBLASTS
CAPILLARY
ENDOTHELIAL CELLS
PERICYTES