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Neutrophils, the major players in healing after myocardial infarction

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Abstract

Neutrophils are a type of immune cells with controversial functions. While their role in the protection against the microbial invasion is well known, the role in controlling and monitoring the non-infection healing processes of the injured tissue and organs are now to be discovered. Particularly in the healing after myocardial infarction, the neutrophils were considered for a long time the "bad guys", sustaining the inflammation and interfering with the repair processes. However, all the attempt to deplete or massively inhibit their activity in experimental conditions or clinical settings lead to catastrophic results, with defective scar formation and worsening of the heart function. As recent studies pointed out the essential role of the neutrophils in monitoring and guiding all the processes involved in the proper tissular healing, a detailed investigation of mechanistic inside is required for a selective and effective design of therapeutical strategies targeting neutrophils to preserve and improve heart function after an acute myocardial infarction.

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Introduction

Neutrophils are a subtype of immune cells responsible for the first line of protection against a microbial infection [1]. They also play the main role in the initiation of a non-infectious inflammatory reaction to repair an injured site or organ [1]. Neutrophils are first cells to come after an acute injury and accumulate massively at the site, inducing and maintaining an inflammatory reaction, forming a granuloma, which will be later removed by the macrophages and replace with scar tissue [2].

Myocardial infarction is the first cause of death in the world, mainly as a consequence of the atherosclerotic blockage of a coronary artery [3]. Despite the intense research of the mechanistic insides in order to find novel therapies, currently, we do not have many options to treat efficiently a myocardial infarction. Thus, even if they survive the critical event, the patients suffering of such a disease will remain with a functional handicap all their life and with an increased risk for a subsequent event, which cannot be prevented. Therefore, the scientific community is looking for viable solutions to reduce the tissular damages after an acute myocardial infarction, and to minimize the consequences on heart biomechanics and function.

As in all other injury events, neutrophils actives immediately and migrate into the infarcted area, in the first seconds/minutes after acute myocardial infarction [3]. Here, they are responsible of creating of a highly inflammatory environment, which was thought to secondary increase the tissular injury on the surrounding cardiomyocytes [4], increasing the infarction size and decreasing the heart function. However, all the clinical trials using anti-inflammatory therapies had catastrophic results [5-8]. Thus, it was clear that the role of neutrophils in the healing after myocardial infarction should be more complex and more detailed the mechanistic investigation involving neutrophils was needed, to be able to create realistic and efficient therapeutic strategies. This review will point out the current knowledge about the role of neutrophils in healing after myocardial infarction and the possible therapeutical strategies which can be employed.

Neutrophils in inflammation

The acute blockage of a coronary artery with the interruption of the blood flow in that region will induce an acute, severe hypoxic injury to the surrounding cardiomyocytes. The damaged cardiomyocytes will secrete multiple inflammatory molecules, inclusive chemokines, which will activate the local cells and recruit immune cells at the injured site to help repair and restoring the tissular integrity [3]. Thus, subsequent tissular healing is a complex process, involving local cells, recruited immune cells and stem cells, which cooperate and interact together to reinstall, as fast as possible, the structural and functional integrity of the heart. Functionally, considering the cell-types and the biological process involved, we can describe this process in 3 different but overlapping phases: the inflammation, the proliferation and the healing phase [3].

Neutrophils seems to be involved in all the phases, controlling and monitoring the succession of events and constitution of the scar [9]. CXCL1/CXCL2 [10], CXCL12/CXCR4 [11] axis, CCR1 [12] are some of the chemotactic molecules involved in the recruitment of neutrophils. Once recruited at the site of the injury, the neutrophils activation will induce their disintegration and release of multitude of inflammatory factors, chemokines and cytokines, danger-molecules or other signaling molecules [13]. Thus, they attract other immune cells, such as inflammatory monocytes [14], which will come shortly after neutrophils, having role of destructing the existent extracellular matrix and cleaning the region of cellular debris [15]. The inflammatory monocytes are another set of immune cells, with inflammatory, proteolytic and phagocytic properties, helping the neutrophils to maintain the inflammatory environment [16]. Despite of the clear role of inducing and sustaining the inflammation, the intend to deplete [17] or reduce [18] the neutrophils after an acute myocardial infarction leads to a defective healing, increase in the infarction size and severe deterioration of the heart function.

Further investigation reveal that neutrophils orchestrate, in fact, the healing process. Impairing this process leads to permanent accumulation of the tissular debris, impairing later the heart function and healing processes [16]. Neutrophils are secreting and releasing the neu-

trophil extracellular traps (NETs) [19, 20], which it is demonstrated to limit the inflammatory reaction to the affected region, trapping the inflammatory mediators inside the area and reducing the spread of the tissular injury into the surrounding areas, contrary to what it was hitherto believed [21, 22].

A selective and specific targeting of neutrophils regarding the inflammatory function seems to have benefic consequences on healing after myocardial infarction and preservation of the heart function [23]. Selective inhibitors/blockers of IL1β reached already phase II in clinical trials [23-25]. Also, a selective inhibition of neutrophils activation, such as CCR3/blockers [26] or phosphatidylserine [27], are demonstrated to inhibit the "bad" side of neutrophils, maintaining their control and monitoring on the healing processes after myocardial infarction.

Neutrophils in proliferation phase

The role of neutrophils in the inflammatory processes is well known, however, the role of neutrophils in the monitoring and coordinating the proliferative and regenerating processes just started to be known [9]. Our preliminary studies reveal that neutrophils are the main cells responsible of stopping the inflammatory processes [17, 26], by inducing the peak of TGF-β1 in fibroblasts. The transitory upregulation of TGF-β1 in myocardium has the role to stop the inflammatory processes and to sustain and increase the anti-inflammatory, pro-fibrotic processes, to fulfil the scar formation and healing after myocardial infarction [28]. TGF-β1 it is an important cytokine with pro-fibrotic role. It is synthetized by many types of cells, such as macrophages and fibroblasts [29]. Once secreted, the TGF-β1 has multiple roles, which are decisive for the proper healing and preserving the heart function [30]. First, the TGF-β1 will stop the inflammatory processes, inducing the polarization of the macrophages towards M2 phenotype [17, 31]. M2 macrophages are known to have a highly anti-inflammatory profile, promoting the angiogenesis and collagen formation [15]. Further, TGF-β1 sustains the differentiation of the fibroblasts into the myofibroblasts [32], which are switching to collagen synthesis under the M2 macrophages [17]. Differentiation towards myofibroblasts will concomitantly inhibiting the release of TGF-β1 from fibroblasts [18], terminating the transitory release and synthesis of this cytokine, also an essential process in healing after myocardial infarction. A prolongation of the TGF-β1 release will increase the fibrotic tissue formation and will maintain the active state of fibroblasts, which will impair later the heart function [18].

The fact that neutrophils are able to control and induce the TGF-β1 transitory synthesis demonstrates the essential role of these cells in the healing after myocardial infarction and explain, in part, the catastrophic results obtained trying to target their function.

Beside the role in monocytes infiltration and fibrosis, neutrophils seem to up-regulate the pro-angiogenic genes in the fibroblasts [33], sustaining the angiogenesis and vessel formation after an acute myocardial infarction [33]. The neutrophils initiate the secretion and accumulation of pro-angiogenic factors at the site of the injury, thus inducing and monitoring the angiogenic processes, which will be sustain and developed later by reparatory monocytes and macrophages.

Neutrophils in scar formation

Extracellular matrix protein synthesis was never investigated in relation with neutrophils, since these cells appeared not to have specific function in this process. However, novel studies pointed out that neutrophils are able to actively interfere with the extracellular matrix synthesis in the fibroblasts [18]. Thus, they can delay the collagen synthesis, upregulating the fibronectin and biglycan [18, 33-35], characteristic for the provisional matrix in the first days after an acute myocardial infarction [36, 37]. This should sustain the transforming processes and integration of fibroblasts to recover the structural integrity of the infarcted regions, avoiding heart rupture and assuring the preservation of the heart function [38, 39].

Targeting the neutrophils has controversial effects on collagen synthesis and scar formation. Thus, targeting CCR1 [12] and CXCR4 [11] receptors reduce neutrophils infiltration, however, no effects was noticed on collagen formation, while depletion of neutrophils induce a chaotic proliferation of fibroblasts, producing large amounts of extracellular matrix proteins, which are not organized accordingly with the re-

quired biomechanics, inducing the severe deterioration of the heart function [17, 18].

Conclusions

Neutrophils are the main cell type initiating, monitoring and controlling all important processes involved in the healing after myocardial infarction. Any therapeutical attempt remained hitherto unsuccessful. Therefore, detailed and sustain investigation is needed to be able to dissect their complex role and to design selective and effective therapeutical strategies to target neutrophils to improve the healing after myocardial infarction.

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Competing interests/conflict of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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