Pericyte Progenitors at the Crossroads Between Fibrosis and Regeneration

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Tissue repair after injury is generally inversely related to the extent of scarring, suggesting the possibility that regeneration could be promoted by interventions that inhibit scar formation. New work by Dulauroy et al has identified a myofibroblast progenitor with pericyte characteristics as an important mediator of scarring in skin and skeletal muscle after injury. The genetic ablation of this lineage, which was identifiable by the expression of ADAM12, led to a dramatic reduction in scarring and more complete regeneration. This work sharpens the conceptual rationale for therapeutic targeting of soluble or cellular mediators of scarring to promote tissue regeneration.

Regenerative medicine has been dominated by the exploration of cell-based therapies, but there are several biological and practical challenges in achieving regeneration with exogenous cell therapies. Despite intense research efforts, the factors dictating and stabilizing progenitor fate in diseased tissues are poorly understood. Without the power to precisely tune and maintain cell fate, the possibility of doing harm is real, particularly with exogenous stem cells that could lead to tumor formation. Endogenous progenitors also have the potential of participating in pathological processes, including fibrotic diseases and cancer. Thus, safety and quality control issues are crucial in the development of cell-based therapies, contributing substantially to potential costs.

In contrast to the explosion in cell therapy approaches, identifying and neutralizing barriers to endogenous regeneration is an understudied approach to achieving tissue repair. A recent study by Dulauroy et al takes such an approach, identifying a myofibroblast progenitor by the expression of the cell membrane-associated protease, ADAM12; this study revealed the progenitor’s contribution to tissue fibrosis after skin and skeletal muscle injury and showed that its genetic ablation prevents scarring (Figure). In this commentary, we discuss how this work clarifies the fate of one category of progenitors residing in the perivascualture, while placing its discovery into the larger context of the interplay between fibrosis and regeneration.

ADAM12 and the Ontogeny of Perivascular Progenitors

Unlike the hematopoietic system, which can be viewed as the prototypical hierarchical cascade with a single multipotent hematopoietic stem cell producing progressively more committed intermediate progenitors, the ontogeny of most tissue-resident progenitors remains incompletely elucidated. Although there is little dispute that most, if not all, tissues contain perivascular cells that exhibit progenitor characteristics, it is increasingly recognized that these progenitors may have diverse developmental origins. Furthermore, the in vivo potency and developmental relationships between such progenitor populations, including pericytes, mesenchymal stem/stromal cells (MSCs), and tissue-specific progenitors, remain relatively unknown.

The genetic lineage tracing experiments performed by Dulauroy et al define a perivascular progenitor in the skin and skeletal muscle, finding that ADAM12 identifies a myofibroblast progenitor lineage arising during development and persisting postnatally with pericyte features. Pericytes are derived from neural crest cells caudal to the aortic arch or from mesothelium for most other tissues below the neck. Potential common developmental origins between ADAM12+ progenitors and pericytes are shown by the expression of ADAM12 by neural crest cells in day 10 mouse embryos and by mesenchymal cells in day 12.5 embryos. Anatomic similarities are found in the uninjured adult skin and muscle, where ADAM12-expressing cells are found in close association with blood vessels, similar to pericytes, which have been historically defined by a shared basement membrane with endothelial cells. More recently, pericytes are increasingly defined by their expression of molecular markers, such as chondroitin sulfate proteoglycan 4 and receptors for platelet-derived growth factors, which are detectable in ADAM12+ cells. These markers are generally not specific for a given lineage or state of commitment; for example, platelet-derived growth factor-α marks a progenitor responsible for ectopic fat formation after injury, whereas ADAM12+ cells, which also express platelet-derived growth factor-α, do not efficiently differentiate into adipocytes. Therefore, despite important molecular and anatomic overlap between ADAM12-expressing progenitors and pericytes, the case for direct ontogenetic links remains largely circumstantial—a familiar quandary in the perivasculature—in large part because the developmental
pathways to the various perivascular progenitor populations have not been definitively described.

In addition to myofibroblasts, lineage-tracing studies have shown in vivo differentiation of pericytes into various terminally differentiated cell types, including adipocytes and myocytes. Viewed collectively, the terminally differentiated products of pericyte lineage-tracing studies functionally overlap with the differentiated progeny of MSCs. MSCs can be isolated from multiple tissues other than bone marrow, and these cells can display multilineage differentiation potential into adipocytes, chondrocytes, osteoblasts, and myocytes. Although the potency of MSCs suggests the possibility of linking several pericyte progenitors with a common ancestor, the existence of MSCs in tissues outside the bone marrow remains controversial because their potency has been largely defined in narrow culture conditions or with in vivo cell transplantation studies that include prolonged ex vivo expansion steps. A specific genetic marker of MSCs remains elusive. Until such a marker is found or new approaches are developed to prospectively track endogenous cell fate in vivo, developmental links between pericyte populations and the existence of a multipotent MSC-like progenitor in the adult perivascularure will remain debated.

Fibrosis Versus Tissue Regeneration
The goal of regenerative medicine is complete tissue repair, which can be considered the restoration of normal tissue architecture and physiological function, without scarring. In this context, the fibroblast becomes an obvious target, given its central role in the generation of scar tissue by the production of extracellular matrix. Although the paradigm of the fibroblast in tissue fibrosis in chronic inflammatory diseases is clear, the role of fibroblasts after acute tissue injury may not always be deleterious. In addition to contributing to the restoration of barrier integrity, fibroblasts assist in orchestrating inflammatory and repair processes by producing signaling cytokines, some of which may promote compensatory healing. Therefore, simply inhibiting scarring may not necessarily translate into improved regeneration.

The demonstration that ADAM12-expressing progenitors were largely responsible for the proinflammatory, connective tissue–producing myofibroblasts after skin and muscle injury provided an avenue to test the hypothesis that inhibition of fibrosis could allow for more robust tissue repair. The most convincing demonstration of this came with the inducible expression of a diphtheria toxin selectively by ADAM12 progenitors, which resulted in their ablation and decreased fibrosis. Coinciding with a quantitative reduction in tissue fibrosis was a qualitative observation of more complete tissue repair. The neutralization of a fibrotic barrier to endogenous cellular repair, however, is not the only potential explanation for the observed regenerative benefit. Indeed, ADAM12 is not just a cell marker, but may also have a direct functional role as a membrane-bound protease involved in transforming growth factor-β and other signaling pathways. Targeted disruption of ADAM12 expression by a specific inhibitory RNA led to similar improvements in inflammatory parameters and fibrosis after muscle injury, corresponding to increased production of proregenerative growth factors. Thus, it is premature to conclude that inhibition of fibrosis is the only mechanism by which myofibroblast neutralization improves regenerative parameters.

Although it may be useful to think of a unidirectional branch point between fibrosis and regeneration, the interplay between these 2 processes likely depends on multiple factors, including endogenous progenitor activity and the ability of the organism to fine-tune the scarring response. Benefits derived from neutralizing myofibroblast progenitors after skin and skeletal muscle injury are likely, in part, attributable to the well-recognized regenerative capacity of these 2 tissues, where, even in the context of scarring, new terminally differentiated effector cells are generated. In tissues characterized by more limited regenerative capacity, such as the brain and heart, simply inhibiting scar formation may not necessarily translate into substantial improvements in cellular repair. Furthermore, even in experimental models where regeneration clearly wins out over tissue fibrosis, as exemplified by the near complete regeneration observed in the zebrafish heart after injury, scar formation may precede complete tissue repair, suggesting that the capacity to abort fibrotic responses and initiate scar resorption may represent additional determinants of the final regenerative outcome.

Figure. Inhibition of scarring as a strategy to promote tissue regeneration. Recent work by Dulauroy et al describes a perivascular ADAM12+ myofibroblast progenitor in skeletal muscle and skin that demonstrates pericyte features, including anatomic association with the basement membrane of blood vessels and expression of pericyte markers. The ablation of myofibroblast progenitors led to a reduction in scarring and more complete tissue repair after muscle or skin injury. This represents one of several conceptual strategies to limit scarring with the ultimate goal of promoting endogenous cell-mediated repair. Other potential approaches are shown in this schematic, including inhibition of myofibroblast production of extracellular matrix and inflammatory cytokines or the stimulation of scar resorption.
**Future Directions**

Existing evidence supports the concept that some perivascular cells are developmental remnants, arrested in a relatively quiescent state before terminal differentiation. Many tissues may contain distal progenitor lineages that are circumstantially linked by their residence in the perivasculature or the expression of pericyte markers. The significance of defining the ontogeny and biology of the perivascular progenitors is emphasized by the remarkable improvements attained by the neutralization of maladaptive myofibroblast progenitors after skin and skeletal muscle injury. Targeting fibrosis by manipulating myofibroblast progenitor activity after injury may represent a therapeutic strategy with broad applicability to tissue regeneration.

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**References**

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