We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,300

Open access books available

172,000

International authors and editors

190M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

Regulatory Mechanisms and Chemical Signaling of Mediators Involved in the Inflammatory Phase of Cutaneous Wound Healing

Fernando Pereira Beserra,
Lucas Fernando Sérgio Gushiken,
Maria Fernanda Hussni and
Cláudia Helena Pellizzon

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/intechopen.81731>

Abstract

Wound healing is a highly complex biological process composed of three overlapping phases: inflammatory, proliferative, and remodeling. The acute inflammatory response has being an integral role in tissue healing and fundamental for the homeostasis and reestablishment. This phase depends on the interaction of cytokines, growth factors, chemokines, and chemical mediators from cells to perform regulatory events and complex interactions of the extracellular matrix, extracellular molecules, soluble mediators, various resident cells such as fibroblasts and keratinocytes, and infiltrated leukocyte subtypes that act to restore or replace the integrity of the skin. If this well-orchestrated response becomes deregulated, the wound can become chronic or progressively fibrotic, with both outcomes impairing tissue function, which can ultimately lead to organ failure and death. In this chapter, we will review the pathway in the skin healing cascade, relating the major chemical inflammatory mediators, cellular and molecular, as well as demonstrating the local and systemic factors that interfere in healing and disorders associated with tissue repair deficiency in chronic inflammations, burns and hypertrophy.

Keywords: wound healing, inflammation, cytokines, growth factors, chemokines

1. Introduction

1.1. Skin wound healing

Skin is the largest organ of all vertebrates, and it is very important to protect the organism against external damage [1]. When the loss of structural integrity of the skin occurs, the organism starts the wound healing process, involving some coordinated, interdependent, and overlapping mechanisms—such as inflammation, cell proliferation, reepithelialization of wounded area, and extra-cellular matrix remodeling—to restructure the skin homeostasis [2, 3]. The initial mechanism of wound healing is the fibrin clot synthesis to avoid bleeding and to keep the local hemostasis, leading to the platelet retention and activation of local vascular mediators [4, 5]. From now on, there is the dilatation of the local vessels due to the release of histamine and serotonin, as well as the increase of vessel permeability, improving the leukocyte migration to the wounded area and starting the inflammatory process. In the first 5 days after the lesion, neutrophils are attracted to the region, removing pathogenic antigens and dead tissue through phagocytosis and protease secretion. After 3 days, there is the macrophage migration to the wounded area, with the maintenance of inflammatory response [5]. Due to the tissue destruction, the local keratinocytes release interleukin-1 β (IL-1 β) and tumor necrosis factor- α (TNF- α), essential cytokines in the inflammatory mechanism, through recruitment and activation of leukocytes in the region and with important roles coordinating other wound healing mechanisms. With these facts, keratinocytes, macrophages, platelets, and endothelial cells of wounded area release some mediators such as growth factors (EGF, FGF, PDGF, TGF- β), cytokines (IL-1 β , IL-6, IL-8, IL-10, TNF- α , IFN- γ), and chemokines, which will control other subsequent mechanisms in skin wound healing [6, 7].

Therefore, the inflammatory mechanism is an important step to the correct and well-coordinated wound healing, modulating the subsequent mechanisms of healing. Furthermore, the comprehension of inflammatory response can lead to new treatments to wound repair and decrease of healing disorders like hypertrophic scars, keloids, chronic inflammation, skin infections, and unwounded lesions [8].

2. Materials and methods

The search for this chapter was carried out on PubMed, Scopus, and Web of Science until June 2018, using “inflammation”, “inflammatory process”, “skin wound healing”, “cytokines”, “chemokines”

Model	Mediator	Target/signaling protein	Biologic effect	References
In vivo BALB/c mice	CD4 cells	IL1 β , IL-6, IL-17, IFN- γ , and CXCL-1 IL-4	The absence of CD4 and CD8 lymphocytes changes in cytokine expression and inflammatory cell infiltrate, but does not influence wound breaking strength, collagen content, or angiogenesis	Chen et al. [50]
	CD8 cells	IL1 β , IL-6, TNF- α , CXCL-1 and CCL-2, IL-4		

Model	Mediator	Target/signaling protein	Biologic effect	References
In vivo BALB/c mice	Neutrophil	MPO, macrophages, and collagens	Neutrophil depletion exhibited significantly accelerated re-epithelialization, without altering the macrophage infiltration or the collagen content in the wound bed	Dovi et al. [51]
In vivo C57BL/6 mice	Epinephrine	IL-6, IL-1 β , TNF- α , IL-1 α , and GM-CSF and PMN	Epinephrine altered the neutrophil (PMN)-dependent inflammatory response to a cutaneous wound through an IL-6 mediated mechanism via β 2 adrenergic receptor-dependent	Kim et al. [52]
In vivo C57Bl/wild-type mice	IL-1 β	Human recombinant IL-1 β , occludin, claudin-1, claudin2, claudin-3, and claudin-5	IL -1 β induced increased claudin -1 expression in cell culture	Rozlomi and Markov [53]
In vivo BALB/C mice	IL-12	Recombinant murine IL-12-rMuIL-12/collagen structure and alignment	IL-12 induced a rapid onset and higher metabolic activity in wounded skin at early time	Li et al. [54]
In vivo BALB/C mice	EFG	EGFR/vaccination extracellular domain (ECD)	Not change the wound healing and inflammatory speed	Fuentes et al. [55]
In vivo BALB/C mice and IL-6 KO mice	IL-6	IL-6/ICAM-1 VCAM-1 IL-1 α IL-1 β MIP-1 α	Delayed angiogenesis and collagen deposition, by the reduced expression of angiogenic and fibrogenic growth factors Reduced inflammatory response	Lin et al. [56]
In vitro Keratinocytes	IL-1 β and TGF- β 1	IL-1 β TGF- β 1/tissue type plasminogen activator (tPa)	IL-1 β interacts com PA by tPA TGF- β 1inibits functional tPA	Lian et al. [57]
In vitro Human and mice fibroblasts	Platelet-derived growth factor (PDGF)	IL-8	Fetal fibroblasts produced less IL-8. Much less IL-8 in stimulated fetal fibroblasts than in adults	Liechty et al. [58]
In vivo C57/Bl6 mice and Mgl2DTR/GFP mice	CD301b macrophage	IL-10, platelet-derived growth factor- β and TGF- β 1	CD301b-expressing subpopulation of macrophages is critical for activation of reparative process	Shook et al. [59]
In vivo Levels and role of chemokine CX3CL1 (fractalkine) and its receptor CX3CR1 in mouse model	CX3CL1 and CX3CR1	MPO, Hydroxyproline (collagen accumulation at the wound sites), TGF-1, VEGF	Inflammation, fibrosis, neovascularization, and regeneration of parenchymal cells were affected by the receptor	Ishida et al. [31]

Model	Mediator	Target/signaling protein	Biologic effect	References
In vivo db/db mice as a diabetic skin wound model.	Peptide inhibitor of complement C1 (PIC1)	complement system (C5), Signal transducer, activator of transcription 4 (STAT4), Leukocyte infiltration, C5a, C3, C3a	PIC1 loaded into the derma CELL did reduce the number of inflammatory cells in the wound bed	Cunnion et al. [60]
In vivo kCYC/mice	IL-10	Mast cells migrating, Macrophages, IL-10, IL-6, aFGF, bFGF, TGF-1, PDGF, TNF- α mRNA	kCYC/mice mast cell increased. IL-10: increased, bFGF decreased in kCYC/mice. IL-10 plays an important role in delayed wound healing	Kimura et al. [61]
In vivo C57BL/6J mice	IL-1 β	p38, MAPK, ERK	IL-1 β stimulates PTGS2 in fibroblast and p38-MAPK in other cells. PGE2 activates INHBA	Arai et al. [62]
In vivo Sprague Dawley Rats	IL-4, IL-12, IL-6, IGF-1 and IFN- α e γ	—	IGF-1, IL-4, IL-6, and IL-3 important in inflammatory phase	Lania et al. [63]
In vivo Mouse model with conditional depletion of macrophages	Inducible diphtheria toxin receptor + diphtheria toxin injections + mice lacking the TGF- β receptor type II (TbRII), Depletion of macrophages	Number of macrophages, neutrophils, and cells positive for activated caspase-3, VEGF-A, or TGF- β 1	Inflammatory phase: reduced formation of vascularized granulation tissue, leading to minimized scar formation. Phase of tissue formation: severe hemorrhage in the wound. Wound closure did not occur. No significant impact in tissue maturation phases.	Lucas et al. [64]
In vivo C57BL/6J mice	Ly6cloMHCIIhi macrophage	IL-17	Ly6cloMHCIIhi macrophages had a non-inflammatory transcriptomic profile and demonstrated that inhibition of IL-17 in mice accelerated normal and delayed healing.	Rodero et al. [65]
In vivo C57BL/6mice	$\gamma\delta$ T Cell	FGF-7, FGF-10, IGF-1, JAML	$\gamma\delta$ T Lymphocytes stimulate the gene and protein expression of important mediators in acute healing model	Xu et al. [66]
Mutant mice ICOS -/- ICOSL -/-	IL-6 and IL-4	—	IL6 and ICOS-ICOSL signaling the skin wound healing in mutant mice	Maeda et al. [67]
In vivo C57BL/6 mice and human acute wounds	Macrophage M1 and M2	IRF-8	IRF-8 is an inflammatory mediator. Inhibition of IRF-8 impairs wound healing.	Guo et al. [68]

Model	Mediator	Target/signaling protein	Biologic effect	References
In vitro Human skin fibroblasts CCD966- SK and HaCaT keratinocytes In vivo BALB/C mice	Fibroblasts and keratinocytes IL-19	IL-19	IRF8 coordinates M1 macrophage population (decrease of M1 mediators IL-1 β , IL-6, TNF- α , iNOS), with no interference in M2 macrophage mediators (arg-1, mrc-1, IL-10) IL-19 upregulates KGF expression in fibroblasts and KGF induces IL-19 expression in keratinocytes KGF promotes keratinocyte proliferation. IL-19 induces keratinocyte migration	Sun et al. [69]
In vivo SKH-1 mice	COX-2	COX-1 and COX-2	Selective inhibition of COX-2 and nonselective inhibition of COX-1 and COX-2 did not affect the healing of sutured surgical incisions in mouse skin.	Blomme et al. [70]
In vivo BALB/c mice	IFN- γ and TGF- β 1	MPO, TGF- β , Stat1, P- Stat1, Smad2, P-Smad2, Smad3, Smad7, α -Tubulin, VEGF, CD3, IL-12p35, IL- 12p40, IL-18, COL1A1	Crosstalk between the IFN- γ /Stat1 and TGF- β 1/Smad signaling pathways in the skin wound healing.	Ishida et al. [71]
In vivo Wistar rats	—	EGF, VEGF, IGF and FGF	Growth factors accelerated the healing process promoting greater angiogenic activity and accelerated fibroplasia and the deposition of type I collagen	de Masi et al. [72]
In vivo C3H/HeJ TLR4- deficient and wild- type C3H/HeOuj mice In vitro NHEK and THP1 cell line	TLR4	CD3+, T cells, Ki67, NF- κ B, p-p38, and p-JNK	TLR4 is activated in early skin wound healing, with a functional mutation of TLR4 results in altered inflammatory cell infiltration, differential cytokine production, and impaired wound closure, besides IL-1 β production by injured keratinocytes is induced through the TLR4- p38/JNK pathway	Chen et al. [73]
In vivo C57BL/6 mice	V γ 4 T cells	IL-17A, IGF-1, CCL20, NF- κ B p65, p-NF- κ B p6, STAT3, p-STAT3, IL-1 β , IL- 23p19	Mechanistic link between V γ 4 T cell-derived IL-17A, epidermal IL-1 β /IL-23, DETC-derived IGF-1, and wound healing responses in the skin	Li et al. [74]

Model	Mediator	Target/signaling protein	Biologic effect	References
In vitro Primary keratinocytes and fibroblasts of IL-6 KO or C57BL/6 mice	IL-6 from keratinocytes and fibroblasts	IL-6, STAT3	IL-6 induces keratinocyte migration indirectly, through the STAT3 activation cascade in fibroblasts, with the synthesis of a fibroblast- derived factor	Gallucci et al. [75]
In vivo Ja18KO (iNKT cell- deficient) mice and C57BL/6 mice	Invariant natural killer T cells (iNKT) and neutrophils	MIP-2, KC, IL-17A, MCP-1, RANTES	MIP-2, KC, IL-17A (neutrophil attractors) were increased in Ja18KO mice. MCP-1 and RANTES (macrophage and lymphocyte attractors) were decreased in Ja18KO mice. Decrease of neutrophil apoptosis in Ja18KO mice. iNKT ciNKT cells promote skin wound healing by regulating neutrophil apoptosis	Tanno et al. [76]
In vivo PPAR- γ KO mice and C57BL/6 J male mice	PPAR- γ from macrophages	PPAR- γ , TNF- α , VEGF, collagen 1	Increase of TNF- α in PPAR- γ KO mice and delay in wound healing	Chen et al. [77]
In vivo Human incision model	IL-4 from mast cells	IL-4, MCP-1	Increased MCP-1 chemoattractant activity in mast cell migration. Increase of IL-4 synthesis by mast cells IL-4 stimulates fibroblast activation	Trautmann et al. [78]
In vivo WBB6F1/J-KitW/ KitW-v mast cell KO female mice and WBB6F1 female mice	Mast cells	TNF- α , MIP-2, VEGF, FGF-2	Decrease of neutrophil infiltration in KO mice. Increase of FGF-2 in KO mice. Mast cells modulate neutrophil infiltration in wound site, with unlikely influence in proliferative phase of wound healing	Egozi et al. [79]
In vivo miR-31 loss-of- function mice	miR-31	NF- κ B, STAT3, RAS/ MAPK	Increase of miR-31 in wound edge keratinocytes during inflammation through NF- κ B and STAT3 pathways miR-31 regulates keratinocyte migration through RAS/MAPK pathway	Shi et al. [80]
In vitro Primary normal human	IL-6, IL-8, TNF- α	STAT3, p38, JNK, EGFR	STAT3, p38, JNK, and NF- κ B activation lead to IL-6, IL-8, and TNF- α increase in	Han et al. [81]

Model	Mediator	Target/signaling protein	Biologic effect	References
keratinocytes In vivo BALB/c female mice			infected wounds. Mixidin2 and mixidin3 modulated inflammatory signaling with anti- inflammatory activity	
In vivo B57BL6/J, B57BL6/ NJ, CD301bGFP- DTR, and Il27Ra ^{-/-} mice	IL-27 (dendritic cells)	Keratin-6 (keratinocytes)	IL-27 is synthesized by dendritic cells and modulates keratinocyte proliferation, migration, and differentiation after skin injury	Yang et al. [82]

Table 1. Chemical mediators involved in the inflammatory response of skin wound healing.

and “growth factors” as keywords. The articles published in the last 20 years were considered (1998–2018). The results are displayed in **Table 1**.

3. Results and discussion

3.1. Mediators involved in the inflammatory phase of wound healing

Wound healing is an extremely dynamic and interactive biological process involving complex interactions of extracellular matrix, extracellular molecules, soluble mediators, multiple resident cells (fibroblasts and keratinocytes), and subtypes of infiltrating leukocytes that together act to restore integrity of the damaged tissue and replace the lost. This process comprises three sequential and overlapping stages, regardless of the amount of injured tissue: hemostasis/inflammation; cell proliferation and matrix repair; and reepithelialization and remodeling of scar tissue, which involve complex biochemical and cellular mechanisms [9].

The inflammatory phase, hemostasis, leukocyte migration, and the beginning of the tissue repair cascade occur. Initially, in response to inflammatory agents, there is reduction of blood flow by vasoconstriction, and with extravasation of blood from the injured vessel, platelets are activated causing the coagulation process to begin [10]. During this process, there is a progressive increase in vascular permeability to migrant cells and biologically active substances. From this process, essential elements for the physiological continuation of healing appear: a fibrin framework, necessary for the migration of the cells that will reach the lesion site, and pro and anti-inflammatory chemo/cytokines that will aid in cell activation and migration [11].

3.2. Neutrophils and macrophages

Neutrophils are the first immune cells recruited into wounded tissue to play a role in reestablishing tissue homeostasis through pathogen phagocytosis and macrophage recruitment as well as excessive neutrophil activity which can contribute to the development of

nonhealing wounds. They play a central role in both killing microbes and promoting wound healing [12]. These cells are extremely important in the inflammatory process, but once recruited into wound sites in such large numbers with exacerbated cytokine secretion, overproduction of reactive oxygen species (ROS), causing extracellular matrix (ECM) and cell membrane damage, and resulting in premature cell senescence [13].

Monocyte-derived macrophages are often considered to be the most important immune cell type in this process. In intact skin, these cells are the most abundant cell types performing sentinel and homeostatic function. The monocytes migrate from vascular circulation to wound. Both infiltrating and resident macrophages on skin are activated by local signals and developed into several subpopulations defined by their different functional phenotypes [14]. Many studies have confirmed that macrophages are critical for proper skin wound healing [15–17]. Upon initial infiltration, proinflammatory macrophages, also called M1, are also responsible for removing cellular debris, damaged matrix, microbes, and neutrophils [17].

3.3. Cytokines

Wound healing is regulated by growth factors and cytokines that are essential not only in the inflammatory process but also in the cell proliferation and maintenance in the repair process by various mechanisms [18].

Cytokines released by neutrophils during apoptosis are chemotactic for monocytes, which start to arrive 5–6 h post injury. IL-1 β is a key interleukin of antimicrobial response by inflammatory response amplification; it stimulates leukocyte recruitment, the release of acute phase proteins, and the increase of permeability of blood vessels [19]. Some authors consider this cytokine as part of a proinflammatory positive feedback loop that sustains a persistent proinflammatory wound macrophage phenotype, contributing to impaired healing of diabetic wounds [18].

TNF- α is a second proinflammatory cytokine that contributes to a chronic wound state. It acts on several stages of leukocyte recruitment mechanism, neutrophils and macrophages, inducing molecular adhesion regulation, chemokine production, and metalloproteinase matrix, as well as tissue inhibitors of metalloproteinases [20]. Interleukin IL-6 is a soluble proinflammatory mediator with pleiotropic activities in inflammation, hematopoiesis, and immune responses [21]. Together with TNF- α and IL-1 β , IL-6 is present in high concentrations in inflammatory processes. After IL-6 is secreted into the area of injury at the beginning of the inflammatory process, it is directed to the liver through the bloodstream, transmitting the information and inducing the hepatocytes to produce certain inflammatory agents [22]. Another important cytokine in the inflammatory phase is IL-8, which also acts as a chemokine (CXCL8). This cytokine is mainly produced by monocytes and in smaller amounts by fibroblasts, endothelial cells, keratinocytes, melanocytes, hepatocytes, and chondrocytes. It usually receives stimuli from other cytokines, such as IL-1, TNF- α , and IFN- γ [23]. The main action of IL-8 is the migration to cells of the immune system, mainly neutrophils, also determining an increase in the expression of endothelial adhesion molecules cells [13].

Since prolonged presence of proinflammatory cytokines may prevent resolution and both pro and anti-inflammatory cytokines are necessary for wound healing, sequential delivery of pro and anti-inflammatory cytokines could be an interesting strategy for improving chronic wound healing [17]. For example, IL-22, considered a proinflammatory cytokine, helped the wound healing of diabetic mice by inducing keratinocyte proliferation and signal transduction and activation of transcription 3 (STAT3) [24]. In this same context but in other tissues, to improve bone repair, decellularized bone was engineered to sequentially release IL-4 (proinflammatory cytokine) and implanted in mice at the site of injury. The sequential release promoted macrophage polarization to switch from a pro to an anti-inflammatory phenotype, resulting in improved wound healing [25].

IL-10 is a regulatory cytokine, which can be secreted by many kinds of immune cells, including Th1, Th2, Th17, Treg, and CD8⁺ T cells, B cells, dendritic cells, macrophages, NK cells, eosinophils, neutrophils, basophils, and MCs, as well as nonimmune cells including keratinocytes. This cytokine is considered an anti-inflammatory cytokine because it is capable of inhibiting the production of other proinflammatory cytokines, such as TNF- α , IL-1 β , and IL-6 [26]. In addition to its potent anti-inflammatory effects, IL-10 has been shown to regulate fibrogenic cytokines, such as transforming growth factor- β (TGF- β), as a part of its role in the regulation of tissue remodeling [27].

3.4. Chemokines

Chemokines are small molecules that induce chemotaxis and activation of certain subsets of leukocytes. They are classified into four types: CC chemokines, CXC chemokines, C chemokines, and CX3C chemokine. Chemokines play important roles in wound healing and are important for maintaining skin homeostasis, and their disruption can result in skin pathologies [28]. They also play important roles in establishing microenvironment in which migratory immune cells, together with skin-resident cells, cause prolonged inflammation [29].

CX3CL1 is expressed by inflamed endothelial cells and epithelial cells, including macrophage, keratinocytes, and vascular smooth muscle cells, whereas CX3CR1 is mainly expressed by neutrophils, monocytes, mast cells, T cells, and NK cells [30]. In the cutaneous wound healing, CX3CL1 has been shown to be expressed by macrophages and endothelial cells, while CX3CR1 is expressed by macrophages and fibroblasts. Decreased expression of macrophage-related cytokines, such as TGF- β and VEGF, and reduced deposition and α -smooth muscle actin and collagen were shown in the injured skin of CX3CR1^{-/-} mice [31].

3.5. Growth factors

Growth factors are naturally occurring endogenous mediators capable of controlling the control of cell growth, proliferation, migration, and differentiation [32]. Once bound specifically to its receptor, the ligand-receptor interaction is able to activate intracellular signal transduction pathways that regulate different cellular functions [33].

PDGF plays a crucial role in the healing process in both chronic and normal wounds. This growth factor is released from degranulating platelets following an injury into the wound fluid [34]. PDGF stimulates mitogenicity and chemotaxis of cells, such as neutrophils, macrophages, fibroblasts, and smooth muscle cells to the site of the wound, initiating the inflammatory process stage [35]. Its function has already been described during the stage of epithelialization of wound healing by upregulating the production of growth factors, such as insulin-like growth factor (IGF)-1 and thrombospondin-1, in turn IGF-1 increases the motility of keratinocyte cells and thrombospondin-1 inhibits proteolytic and enzymatic degradation of PDGF [36].

Angiogenesis is an extremely important process in normal development and tissue homeostasis and repair, besides contributes directly also to various forms of pathology, such as tumor development and metastasis, psoriasis, rheumatoid arthritis, and wet macular degeneration [37]. One of the most important proangiogenic mediators is vascular endothelial growth factor (VEGF), responsible for stimulating new blood vessels formation, tissue proliferation, migration, differentiation, and survival, which contribute to the angiogenesis process, in addition to influencing the repair and wound closure and granulation tissue formation [38]. The VEGF family has several members, and one of its members such as VEGF-A begins the process of wound healing promoting biological events linked to angiogenesis and migration of endothelial cells [39]. Administration of VEGF-A has been reported to restore impairment of angiogenesis in diabetic ischemic limbs in an animal model as well as to improve the reepithelialization process of diabetic wounds [40].

Epidermal growth factor (EGF) stimulates proliferation and differentiation of various cells, including fibroblasts, endothelial cells, and epithelial cells, and shows mitogenic and migratory activity on the edge keratinocytes of the lesions [41]. EGF participates in this mechanism, which is considered essential in the cutaneous wound healing, which begins a few hours after the injury, but presents a more evident activity in the proliferative phase of wound healing, and continues until the extracellular matrix remodeling phase [42].

Another growth factor that involves healing process activity is the family of fibroblast growth factors (FGF), which have already been reported to play crucial events in the wound healing process [43]. FGFs are secreted by keratinocytes, fibroblasts, endothelial cells, smooth muscle cells, chondrocytes, and mast cells [44]. During an acute cutaneous wound process, it has been reported an increase in the production of FGF-2 and that they are responsible for formation of granulation tissue, reepithelialization, and tissue remodeling [45]. Moreover, functions such as synthesis, deposition of various constituents of the extracellular matrix, and increased motility of keratinocytes are regulated by FGF-2 [46].

Transforming growth factor type- β (TGF- β) activity in the healing process was analyzed by being one of the proteins with the greatest spectrum of activities, with effect on cell proliferation, differentiation and production of extracellular matrix, and immunological modulation [46, 47]. Moreover, TGF- β has many biological activities and is thought to be a particularly important contributor to fibrosis, angiogenesis, and tissue repair. This growth factor can also influence T cells, including Th17 and Treg cells, as well as B cells, dendritic cells, NK cells, neutrophils, and eosinophils [48, 49].

4. Conclusion

Increasing scientific knowledge has contributed to define highly coordinated molecular and cellular events involved in the cutaneous wound healing. Recent findings show that there is a clear correlation between the stage of the wound and its effectiveness in the healing process, and endogenous mediators, such as cytokines, chemokines, and growth factors, indicate important and crucial steps in the normal healing and are useful as prognostic indicators. Although much is known about the cellular and molecular basis of normal skin healing, there are still avenues of research left to unravel that will guide us to better therapies, new therapeutic targets, and strategies for the skin wound treatment, especially chronic wounds.

Conflict of interest

The authors declare no conflict of interests.

Author details

Fernando Pereira Beserra¹, Lucas Fernando Sérgio Gushiken¹, Maria Fernanda Hussni² and Cláudia Helena Pellizzon^{1*}

*Address all correspondence to: claudia@ibb.unesp.br

1 Department of Morphology, Institute of Biosciences of Botucatu, São Paulo State University (UNESP), São Paulo, Brazil

2 Undergraduate Student School of Veterinary Medicine and Animal Science, São Paulo State University (UNESP), São Paulo, Brazil

References

- [1] Fikru A, Makonnen E, Eguale T, Debella A, Mekonnen GA. Evaluation of in vivo wound healing activity of methanol extract of *Achyranthes aspera* L. Journal of Ethnopharmacology. 2012;**143**:469-474. DOI: 10.1016/j.jep.2012.06.049
- [2] Martins VL, Caley M, O'Toole EA. Matrix metalloproteinases and epidermal wound repair. Cell and Tissue Research. 2013;**351**:255-268. DOI: 10.1007/s00441-012-1410-z
- [3] Agha R, Ogawa R, Pietramaggiori G, Orgill DP. A review of the role of mechanical forces in cutaneous wound healing. The Journal of Surgical Research. 2011;**171**:700-708. DOI: 10.1016/j.jss.2011.07.007

- [4] Gurtner GC, Werner S, Barrandon Y, Longaker MT. Wound repair and regeneration. *Nature*. 2008;**453**:314-321. DOI: 10.1038/nature07039
- [5] Tziotzios C, Profyris C, Sterling J. Cutaneous scarring: Pathophysiology, molecular mechanisms, and scar reduction therapeutics. *Journal of the American Academy of Dermatology*. 2012;**66**:13-24. DOI: 10.1016/j.jaad.2011.08.035
- [6] Reinke JM, Sorg H. Wound repair and regeneration. *European Surgical Research*. 2012;**49**:35-43. DOI: 10.1159/000339613
- [7] Singer AJ, Clark RAF. Cutaneous wound healing. *The New England Journal of Medicine*. 1999;**341**:738-746. DOI: 10.1056/NEJM199909023411006
- [8] Eming SA, Krieg T, Davidson JM. Inflammation in wound repair: Molecular and cellular mechanisms. *The Journal of Investigative Dermatology*. 2017;**127**:514-525. DOI: 10.1038/sj.jid.5700701
- [9] Boateng JS, Matthews KH, Stevens HNE, Eccleston GM. Wound healing dressings and drug delivery systems: A review. *Journal of Pharmaceutical Sciences*. 2008;**97**:2892-2923. DOI: 10.1002/jps.21210
- [10] Martin P, Feng Y. Inflammation: Wound healing in zebrafish. *Nature*. 2009;**459**:921-923. DOI: 10.1038/459921a
- [11] Morin C, Roumegous A, Carpentier G, Barbier-Chassefière V, Garrigue-Antar L, Careda S, et al. Modulation of inflammation by Cicaderma ointment accelerates skin wound healing. *The Journal of Pharmacology and Experimental Therapeutics*. 2012;**343**:115-124. DOI: 10.1124/jpet.111.188599
- [12] Wilgus TA, Roy S, McDaniel JC. Neutrophils and wound repair: Positive actions and negative reactions. *Advances in Wound Care*. 2013;**2**:379-388. DOI: 10.1089/wound.2012.0383
- [13] Kolaczowska E, Kubes P. Neutrophil recruitment and function in health and inflammation. *Nature Reviews. Immunology*. 2013;**13**:159-175. DOI: 10.1038/nri3399
- [14] Murray PJ, Wynn TA. Protective and pathogenic functions of macrophage subsets. *Nature Reviews. Immunology*. 2012;**11**:723-737. DOI: 10.1038/nri3073.Protective
- [15] Nagaoka T, Kaburagi Y, Hamaguchi Y, Hasegawa M, Takehara K, Steeber DA, et al. Delayed wound healing in the absence of intercellular adhesion molecule-1 or L-selectin expression. *The American Journal of Pathology*. 2000;**157**:237-247. DOI: 10.1016/S0002-9440(10)64534-8
- [16] Mori R, Kondo T, Ohshima T, Ishida Y, Mukaida N. Accelerated wound healing in tumor necrosis factor receptor p55-deficient mice with reduced leukocyte infiltration. *The FASEB Journal*. 2002;**16**:963-974
- [17] Larouche J, Sheoran S, Maruyama K, Martino MM. Immune regulation of skin wound healing: Mechanisms and novel therapeutic targets. *Advances in Wound Care*. 2018;**7**(7):209-231. DOI: 10.1089/wound.2017.0761

- [18] Zhao R, Liang H, Clarke E, Jackson C, Xue M. Inflammation in chronic wounds. *International Journal of Molecular Sciences*. 2016;**17**:1-14. DOI: 10.3390/ijms17122085
- [19] MacLeod AS, Rudolph R, Corriden R, Ye I, Garijo O, Havran WL. Skin-resident T cells sense ultraviolet radiation-induced injury and contribute to DNA repair. *Journal of Immunology*. 2014;**192**:5695-5702. DOI: 10.4049/jimmunol.1303297
- [20] Gragnani A, Müller BR, da Silva IDC, de Noronha SMR, Ferreira LM. Keratinocyte growth factor, tumor necrosis factor- α and interleukin-1 β gene expression in cultured fibroblasts and keratinocytes from burned patients. *Acta Cirurgica Brasileira*. 2013;**28**:551-558. DOI: 10.1590/s1808-98762013000300003
- [21] Tanaka T, Narazaki M, Kishimoto T. IL-6 in inflammation, immunity, and disease. *Cold Spring Harbor Perspectives in Biology*. 2014;**6**:a016295
- [22] Scheller J, Chalaris A, Schmidt-arras D, Rose-john S. The pro- and anti-inflammatory properties of the cytokine interleukin-6. *Biochimica et Biophysica Acta*. 2011;**1813**:878-888. DOI: 10.1016/j.bbamcr.2011.01.034
- [23] Mayadas TN, Cullere X, Lowell CA. The multifaceted functions of neutrophils. *Annual Review of Pathology Mechanisms of Disease*. 2014;**9**:181-218. DOI: 10.1146/annurev-pathol-020712-164023
- [24] Avitabile S, Odorisio T, Madonna S, Eyerich S, Guerra L, Eyerich K, et al. Interleukin-22 promotes wound repair in diabetes by improving keratinocyte pro-healing functions. *The Journal of Investigative Dermatology*. 2015;**135**:2862-2870. DOI: 10.1038/jid.2015.278
- [25] Spiller KL, Nassiri S, Witherell CE, Anfang RR, Ng J, Nakazawa KR, et al. Sequential delivery of immunomodulatory cytokines to facilitate the M1-to-M2 transition of macrophages and enhance vascularization of bone scaffolds. *Biomaterials*. 2015;**37**:194-207. DOI: 10.1016/j.biomaterials.2014.10.017
- [26] Saraiva M, O'Garra A. The regulation of IL-10 production by immune cells. *Nature Reviews. Immunology*. 2010;**10**:170-181. DOI: 10.1038/nri2711
- [27] Yamamoto T, Eckes B, Krieg T. Effect of Interleukin-10 on the gene expression of type I collagen, fibronectin, and decorin in human skin fibroblasts: Differential regulation by transforming growth factor- β and monocyte chemoattractant protein-1. *Biochemical and Biophysical Research Communications*. 2001;**281**:200-205. DOI: 10.1006/bbrc.2001.4321
- [28] Nakayama T, Watanabe Y, Oiso N, Higuchi T, Shigeta A, Mizuguchi N, et al. Eotaxin-3/CC chemokine ligand 26 is a functional ligand for CX3CR1. *Journal of Immunology*. 2010;**185**:6472-6479. DOI: 10.4049/jimmunol.0904126
- [29] Sugaya M. Chemokines and skin diseases. *Archivum Immunologiae et Therapiae Experimentalis (Warsz)*. 2015;**63**:109-115. DOI: 10.1007/s00005-014-0313-y
- [30] White GE, Greaves DR. Fractalkine: A survivor's guide chemokines as antiapoptotic mediators. *Arteriosclerosis, Thrombosis, and Vascular Biology*. 2012;**32**:589-594. DOI: 10.1161/ATVBAHA.111.237412

- [31] Ishida Y, Gao J-L, Murphy PM. Chemokine receptor CX3CR1 mediates skin wound healing by promoting macrophage and fibroblast accumulation and function. *Journal of Immunology*. 2008;**180**:569-579. DOI: 10.4049/jimmunol.180.1.569
- [32] Barrientos S, Stojadinovic O, Golinko MS, Brem H, Tomic-Canic M. Growth factors and cytokines in wound healing. *Wound Repair and Regeneration*. 2008;**16**:585-601. DOI: 10.1111/j.1524-475X.2008.00410.x
- [33] Koria P. Delivery of growth factors for tissue regeneration and wound healing. *BioDrugs*. 2012;**26**:163-175. DOI: 10.2165/11631850-000000000-00000
- [34] Uysal AC, Mizuno H, Tobita M, Ogawa R, Hyakusoku H. The effect of adipose-derived stem cells on ischemia-reperfusion injury: Immunohistochemical and ultrastructural evaluation. *Plastic and Reconstructive Surgery*. 2009;**124**:804-815. DOI: 10.1097/PRS.0b013e3181b17bb4
- [35] Mardani M, Asadi-Samani M, Rezapour S, Rezapour P. Evaluation of bred fish and seawater fish in terms of nutritional value, and heavy metals. *Journal of Chemical and Pharmaceutical Sciences*. 2016;**9**:1277-1283
- [36] Mardani M, Mahmoud B, Moradmand JS, Salehi A, Davoodi M, Ghobadi S, et al. Comparison of the descurainia sophia and levostatin effect on the ldl cholesterol reduction, a clinical trial study. *Journal of Chemical and Pharmaceutical Sciences*. 2016;**9**:1329-1333
- [37] Nagy JA, Dvorak AM, Dvorak HF. Vascular hyperpermeability, angiogenesis, and stroma generation. *Cold Spring Harbor Perspectives in Medicine*. 2012;**2**:a006544. DOI: 10.1101/cshperspect.a006544
- [38] Kim HR, Lee JH, Kim KW, Kim BM, Lee SH. The relationship between synovial fluid VEGF and serum leptin with ultrasonographic findings in knee osteoarthritis. *International Journal of Rheumatic Diseases*. 2016;**19**:233-240. DOI: 10.1111/1756-185X.12486
- [39] Huang SP, Hsu CC, Chang SC, Wang CH, Deng SC, Dai NT, et al. Adipose-derived stem cells seeded on acellular dermal matrix grafts enhance wound healing in a murine model of a full-thickness defect. *Annals of Plastic Surgery*. 2012;**69**:656-662. DOI: 10.1097/SAP.0b013e318273f909
- [40] Song SH, Lee MO, Lee JS, Jeong HC, Kim HG, Kim WS, et al. Genetic modification of human adipose-derived stem cells for promoting wound healing. *Journal of Dermatological Science*. 2012;**66**:98-107. DOI: 10.1016/j.jdermsci.2012.02.010
- [41] Singh B, Carpenter G, Coffey RJ. EGF receptor ligands: Recent advances. *F1000Research*. 2016;**5**:2270. DOI: 10.12688/f1000research.9025.1
- [42] Werner S, Grose R. Regulation of wound healing by growth factors and cytokines. *Physiological Reviews*. 2003;**83**:835-870
- [43] Ghafarzadeh M, Eatemadi A. Clinical efficacy of liposome-encapsulated Aloe vera on melasma treatment during pregnancy. *Journal of Cosmetic and Laser Therapy*. 2017;**19**:181-187. DOI: 10.1080/14764172.2017.1279329

- [44] Nie C, Yang D, Xu J, Si Z, Jin X, Zhang J. Locally administered adipose-derived stem cells accelerate wound healing through differentiation and vasculogenesis. *Cell Transplantation*. 2011;**20**:205-216. DOI: 10.3727/096368910X520065
- [45] Jaferian S, Soleymaninejad M, Negahdari B, Eatemadi A. Stem cell, biomaterials and growth factors therapy for hepatocellular carcinoma. *Biomedicine and Pharmacotherapy*. 2017;**88**:1046-1053. DOI: 10.1016/j.biopha.2017.01.154
- [46] Lee SH, Lee JH, Cho KH. Effects of human adipose-derived stem cells on cutaneous wound healing in nude mice. *Annals of Dermatology*. 2011;**23**:150-155. DOI: 10.5021/ad.2011.23.2.150
- [47] Leask A, Abraham DJ. TGF-beta signaling and the fibrotic response. *The FASEB Journal*. 2004;**18**:816-827. DOI: 10.1096/fj.03-1273rev
- [48] Biernacka A, Dobaczewski M, Frangogiannis NG. TGF-b signaling in fibrosis. *Growth Factors*. 2011;**29**:196-202. DOI: 10.3109/08977194.2011.595714
- [49] Sanjabi S, Oh SA, Li MO. Regulation of the immune response by TGF-B: From conception to autoimmunity and infection. *Cold Spring Harbor Perspectives in Biology*. 2017;**9**:a022236. DOI: 10.1101/cshperspect.a022236
- [50] Chen L, Mehta ND, Zhao Y, DiPietro LA. Absence of CD4 or CD8 lymphocytes changes infiltration of inflammatory cells and profiles of cytokine expression in skin wounds, but does not impair healing. *Experimental Dermatology*. 2014;**23**:189-194. DOI: 10.1021/nn300902w. Release
- [51] Dovi JV, He LK, DiPietro L. Accelerated wound closure in neutrophil-depleted mice. *Journal of Leukocyte Biology*. 2003;**73**:448-455. DOI: 10.1189/jlb.0802406
- [52] Kim MH, Gorouhi F, Ramirez S, Granick JL, Byrne BA, Soulika AM, et al. Catecholamine stress alters neutrophils trafficking and impairs wound healing by beta2 adrenergic receptor mediated upregulation of IL-6. *The Journal of Investigative Dermatology*. 2014;**134**:809-817. DOI: 10.1038/jid.2013.415.Catecholamine
- [53] Rozlomiy VL, Markov AG. Effect of interleukin-1beta on the expression of tight junction proteins in the culture of HaCaT keratinocytes. *Bulletin of Experimental Biology and Medicine*. 2010;**149**:280-283
- [54] Li J, Bower AJ, Vainstein V, Gluzman-Poltorak Z, Chaney EJ, Marjanovic M, et al. Effect of recombinant interleukin-12 on murine skin regeneration and cell dynamics using in vivo multimodal microscopy. *Biomedical Optics Express*. 2015;**6**:4277. DOI: 10.1364/BOE.6.004277
- [55] Fuentes D, Chacón L, Casacó A, Ledón N, Fernández N, Iglesias A, et al. Effects of an epidermal growth factor receptor-based cancer vaccine on wound healing and inflammation processes in murine experimental models. *International Wound Journal*. 2014;**11**:98-103. DOI: 10.1111/j.1742-481X.2012.01074.x
- [56] Lin ZQ. Essential involvement of IL-6 in the skin wound-healing process as evidenced by delayed wound healing in IL-6-deficient mice. *Journal of Leukocyte Biology*. 2003;**73**:713-721. DOI: 10.1189/jlb.0802397

- [57] Lian X, Yang L, Gao Q, Yang T. IL-1 α is a potent stimulator of keratinocyte tissue plasminogen activator expression and regulated by TGF- β 1. *Archives of Dermatological Research*. 2008;**300**:185-193. DOI: 10.1007/s00403-007-0828-8
- [58] Liechty KW, Crombleholme TM, Cass DL, Martin B, Adzick NS. Diminished interleukin-8 (IL-8) production in the fetal wound healing response. *The Journal of Surgical Research*. 1998;**77**:80-84. DOI: 10.1006/jsre.1998.5345
- [59] Shook B, Xiao E, Kumamoto Y, Iwasaki A, Horsley V. CD301b+ macrophages are essential for effective skin wound healing. *The Journal of Investigative Dermatology*. 2016;**136**:1885-1891. DOI: 10.1038/ng.3641.Punctuated
- [60] Cunnion KM, Krishna NK, Pallera HK, Pinos-Fernandez A, Rivera MG, Hair PS, et al. Complement activation and STAT4 expression are associated with early inflammation in diabetic wounds. *PLoS One*. 2017;**12**:1-19. DOI: 10.1371/journal.pone.0170500
- [61] Kimura T, Sugaya M, Blauvelt A, Okochi H, Sato S. Delayed wound healing due to increased interleukin-10 expression in mice with lymphatic dysfunction. *Journal of Leukocyte Biology*. 2013;**94**:137-145. DOI: 10.1189/jlb.0812408
- [62] Arai KY, Ono M, Kudo C, Fujioka A, Okamura R, Nomura Y, et al. IL-1 β stimulates activin β A mRNA expression in human skin fibroblasts through the MAPK pathways, the nuclear factor- κ B pathway, and prostaglandin E2. *Endocrinology*. 2011;**152**:3779-3790. DOI: 10.1210/en.2011-0255
- [63] Lania BG, Morari J, De Souza AL, Da Silva MN, De Almeida AR, Veira-Damiani G, et al. Topical use and systemic action of green and roasted coffee oils and ground oils in a cutaneous incision model in rats (*Rattus norvegicus albinus*). *PLoS One*. 2017;**12**:1-17. DOI: 10.1371/journal.pone.0188779
- [64] Lucas T, Waisman A, Ranjan R, Roes J, Krieg T, Muller W, et al. Differential roles of macrophages in diverse phases of skin repair. *Journal of Immunology*. 2010;**184**:3964-3977. DOI: 10.4049/jimmunol.0903356
- [65] Rodero MP, Hodgson SS, Hollier B, Combadiere C, Khosrotehrani K. Reduced IL-17a expression distinguishes a Ly6c lo MHCII^{hi} macrophage population promoting wound healing. *The Journal of Investigative Dermatology*. 2013;**133**:783-792. DOI: 10.1038/jid.2012.368
- [66] Xu P, Fu X, Xiao N, Guo Y, Pei Q, Peng Y, et al. Involvements of $\gamma\delta$ T lymphocytes in acute and chronic skin wound repair. *Inflammation*. 2017;**40**:1416-1427. DOI: 10.1007/s10753-017-0585-6
- [67] Maeda S, Fujimoto M, Matsushita T, Hamaguchi Y, Takehara K, Hasegawa M. Inducible costimulator (ICOS) and ICOS ligand signaling has pivotal roles in skin wound healing via cytokine production. *The American Journal of Pathology*. 2011;**179**:2360-2369. DOI: 10.1016/j.ajpath.2011.07.048

- [68] Guo Y, Yang Z, Wu S, Xu P, Peng Y, Yao M. Inhibition of IRF8 negatively regulates macrophage function and impairs cutaneous wound healing. *Inflammation*. 2017;**40**: 68-78. DOI: 10.1007/s10753-016-0454-8
- [69] Sun D, Yeh C-H, So E, Wang L, Wei T, Chang M, et al. Interleukin (IL)-19 promoted skin wound healing by increasing fibroblast keratinocyte growth factor expression. *Cytokine*. 2013;**62**:360-368. DOI: 10.1016/j.cyto.2013.03.017
- [70] Blomme EAG, Chinn KS, Hardy MM, Casler JJ, Kim SH, Opsahl AC, et al. Selective cyclooxygenase-2 inhibition does not affect the healing of cutaneous full-thickness incisional wounds in SKH-1 mice. *The British Journal of Dermatology*. 2003;**148**:211-223. DOI: 10.1046/j.1365-2133.2003.05065.x
- [71] Ishida Y, Kondo T, Takayasu T, Iwakura Y, Mukaida N. The essential involvement of cross-talk between IFN- and TGF- in the skin wound-healing process. *Journal of Immunology*. 2004;**172**:1848-1855. DOI: 10.4049/jimmunol.172.3.1848
- [72] De Masi ECDJ, Campos ACL, De Masi FDJ, Ratti MAS, Ike IS, De Masi RDJ. A influência de fatores de crescimento na cicatrização de feridas cutâneas de ratas. *Brazilian Journal of Otorhinolaryngology*. 2016;**82**:512-521. DOI: 10.1016/j.bjorl.2015.09.011
- [73] Chen L, Guo S, Ranzer MJ, Dipietro LA. Toll-like receptor 4 plays an essential role in early skin wound healing. *Journal of Investigative Dermatology*. 2013;**133**:258-267. DOI: 10.1038/jid.2012.267.Toll-like
- [74] Li Y, Wang Y, Zhou L, Liu M, Liang G, Yan R, et al. V γ 4 T cells inhibit the pro-healing functions of dendritic epidermal T cells to delay skin wound closure through IL-17A. *Frontiers in Immunology*. 2018;**9**:1-19. DOI: 10.3389/fimmu.2018.00240
- [75] Gallucci RM, Sloan DK, Heck JM, Murray AR, O'Dell SJ. Interleukin 6 indirectly induces keratinocyte migration. *The Journal of Investigative Dermatology*. 2004;**122**:764-772. DOI: 10.1111/j.0022-202X.2004.22323.x
- [76] Tanno H, Kawakami K, Kanno E, Suzuki A, Takagi N, Yamamoto H, et al. Invariant NKT cells promote skin wound healing by preventing a prolonged neutrophilic inflammatory response. *Wound Repair and Regeneration*. 2017;**25**:805-815. DOI: 10.1111/wrr.12588
- [77] Chen H, Shi R, Luo B, Yang X, Qiu L, Xiong J, et al. Macrophage peroxisome proliferator-activated receptor B deficiency delays skin wound healing through impairing apoptotic cell clearance in mice. *Cell Death and Disease*. 2015;**6**:1-13. DOI: 10.1038/cddis.2014.544
- [78] Trautmann A, Toksoy A, Engelhardt E, Bröcker E-B, Gillitzer R. Mast cell involvement in normal human skin wound healing: Expression of monocyte chemoattractant protein-1 is correlated with recruitment of mast cells which synthesize interleukin-4 in vivo. *The Journal of Pathology*. 2000;**190**:100-106. DOI: 10.1002/(SICI)1096-9896(200001)190:1<100::AID-PATH496>3.0.CO;2-Q

- [79] Egozi EI, Ferreira AM, Burns AL, Gamelli RL, Dipietro LA. Mast cells modulate the inflammatory but not the proliferative response in healing wounds. *Wound Repair and Regeneration*. 2003;**11**:46-54. DOI: 10.1046/j.1524-475X.2003.11108.x
- [80] Shi J, Ma X, Su Y, Song Y, Tian Y, Yuan S, et al. MiR-31 mediates inflammatory signaling to promote re-epithelialization during skin wound healing. *The Journal of Investigative Dermatology*. 2018;**18**:31857-31858. DOI: 10.1016/j.jid.2018.03.1521
- [81] Han HM, Ko S, Cheong MJ, Bang JK, Seo CH, Luchian T, et al. Myxinidin2 and myxinidin3 suppress inflammatory responses through STAT3 and MAPKs to promote wound healing. *Oncotarget*. 2017;**8**:87582-87597. DOI: 10.18632/oncotarget.20908
- [82] Yang B, Suwanpradid J, Sanches-Lagunes R, Choi HW, Hoang P, Wang D, et al. IL-27 facilitates skin wound healing through induction of epidermal proliferation and host defense. *The Journal of Investigative Dermatology*. 2017;**137**:1166-1175. DOI: 10.1016/j.clinbiochem.2015.06.023. Gut-Liver