Available online at www.sciencedirect.com

SCIENCE DIRECT®

Biochimica et Biophysica Acta 1711 (2005) 197–207

BIOCHIMICA ET BIOPHYSICA ACTA
BBA

http://www.elsevier.com/locate/bba

Review

Gap junctional communication in tissue inflammation and repair

Marc Chanson[a,*], Jean-Paul Derouette[b], Isabelle Roth[b], Bernard Foglia[a,b], Isabelle Scerri[a], Tecla Dudez[a], Brenda R. Kwak[b]

[a]*Laboratory of Clinical Investigation III, Department of Pediatrics, HUG-P.O. BOX 14, Micheli-du-Crest, 24, 1211 Geneva 14, Switzerland*
[b]*Division of Cardiology, Geneva University Hospitals, Geneva, Switzerland*

Received 11 June 2004; received in revised form 12 October 2004; accepted 14 October 2004
Available online 30 October 2004

Abstract

Local injury induces a complex orchestrated response to stimulate healing of injured tissues, cellular regeneration and phagocytosis. Practically, inflammation is defined as a defense process whereby fluid and white blood cells accumulate at a site of injury. The balance of cytokines, chemokines, and growth factors is likely to play a key role in regulating important cell functions such as migration, proliferation, and matrix synthesis during the process of inflammation. Hence, the initiation, maintenance, and resolution of innate responses depend upon cellular communication. A process similar to tissue repair and subsequent scarring is found in a variety of fibrotic diseases. This may occur in a single organ such as liver, kidneys, pancreas, lung, skin, and heart, but fibrosis may also have a more generalized distribution such as in atherosclerosis. The purpose of this review is to summarize recent advances on the contribution of gap junction-mediated intercellular communication in the modulation of the inflammatory response and tissue repair.

Keywords: Gap junction; Connexin; Inflammation; Wound healing; Tissue repair

Contents

1. Tissue homeostasis

The sequence of events involved in maintenance of tissue homeostasis encompasses mechanisms within single cells as well as interactions between cells within a population. Among structures such as desmosomes, tight junctions, and adherens junctions, gap junctions are thought to play crucial roles in these interactions by contributing to the ability of cells to share cytoplasmic components, a process referred to as gap junctional intercellular communication (GJIC) [1–5].

The analyses of the three-dimensional structure of gap junctions and their constituent channels have been significantly elucidated in vertebrates. Gap junctions consist of aggregates of transmembrane hemichannels (or connexons) that dock to similar connexons on the neighboring cell. Three-dimensional structure of a recombinant gap junction

* Corresponding author. Tel.: +41 22 37 24 611; fax: +41 22 37 24 088.
E-mail address: Marc.Chanson@hcuge.ch (M. Chanson).

0005-2736/$ - see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.bbamem.2004.10.005

channel revealed that opposing connexons are staggered by 30° and packed in the intercellular gap [6,7], resulting in a tight seal between the two hemichannels. The wall of each connexon is formed of six protein subunits, termed connexins (Cx), creating a 2-nm-diameter aqueous pore that allows diffusion of molecules of about 1000 Da between the cytoplasm of adjacent cells [1–5]. The connexin channels act as molecular sieves, allowing the direct transfer of ions, small molecules, and second messengers. An important role of GJIC is to transmit information on the cell's functional state to its neighbors, a property of gap junction channels that varies with their connexin composition. With the sequencing of the human genome, the connexins are now known to be members of a family of at least 20 proteins [8]. Most tissues express more than one connexin type that can be regulated by hormones, growth factors, and proinflammatory mediators, thus ensuring a fine-tuned regulation of GJIC [3,4,9–12].

Gap junctions drive numerous important biological processes such as the rapid transmission of electrotonic signals to coordinate contraction of cardiac and smooth muscles, the intercellular propagation of Ca^{2+} waves and the transfer of signals to enable proper embryogenesis. Tissue homeostasis depends not simply on intercellular communication but also on the specific pattern of connexin gene expression [13–16]. Thus, changes in cell coupling resulting in tissue dysfunction and pathological conditions are associated with altered pattern of connexin expression and/or GJIC regulation. The regulation of connexin expression is effected at different levels from transcription to post-translational processes, although the mechanisms involved are poorly defined.

Critical functions for gap junction channels have been elucidated by the discovery of disease-causing mutations in human connexin genes and the observation that mice with targeted deletions of connexins develop distinct phenotypes [8,17]. Mutations in connexins or defective production of gap junctions in humans are associated with several diseases, including deafness, Charcot-Marie-Tooth X-linked neuropathy, cataractogenesis and a variety of skin disorders [18–20]. Altogether, these recent findings confirm the view that perturbation of gap junction-mediated homeostasis contributes to disease initiation and/or progression. The underlying mechanisms by which GJIC contributes and/or modulates tissue injury are, however, unknown. Here, we will discuss recent advances on the contribution of gap junction-mediated intercellular communication in the modulation of the inflammatory response and tissue repair. Detailed reviews on possible roles for gap junctions in tissue injury and in inflammatory and immune cells recently appeared [21–24]. Because a chapter has already been dedicated to gap junctions in neurological disorders of the central nervous systems in the first special issue from this journal [25], the role of GJIC in brain damage/inflammation will not be further treated here.

2. Tissue inflammation

The repair of tissue damage is a survival process that involves cell–cell and cell–matrix interactions. Within minutes after injury and loss of tissue homeostasis, cytokines and chemokines are released from blood, injured tissues, or parenchymal cells (Fig. 1). Gap junction channel

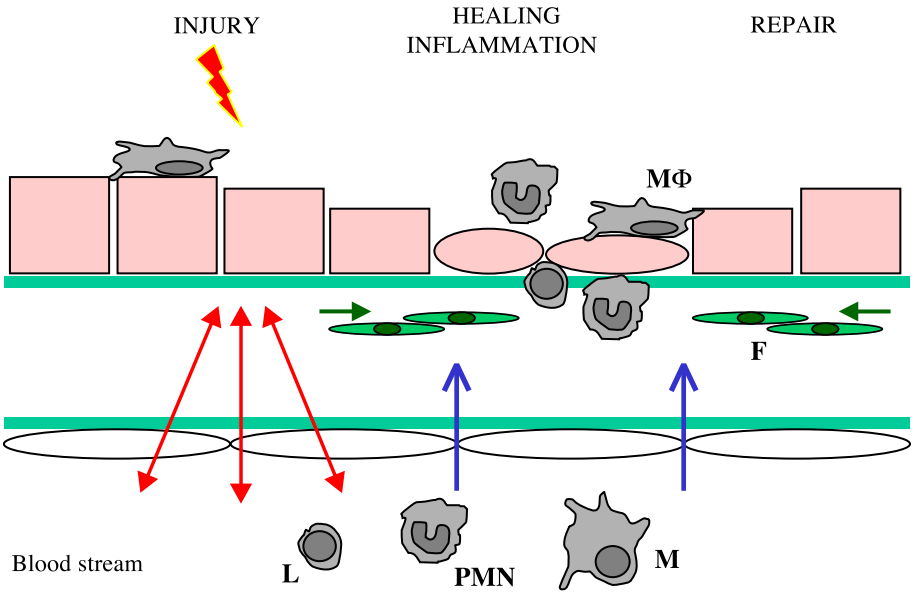

Fig. 1. Representative view of the sequence of events occurring from cell damage to tissue repair. Following injury or infection, cytokines and chemokines are released from blood, injured tissues, or parenchymal cells (red arrowheads). This production rapidly stimulates leukocyte extravasations and migration into tissues (blue arrows). The first blood cell members to appear are polymorphomononuclear cells (PMNs) of which the most abundant are neutrophils, followed by monocytes (M) that differentiate into macrophages (MΦ). Finally, the immune response is completed with the immigration of lymphocytes (L). The production of cytokines and of a variety of growth factors by the damaged tissue also triggers the migration of resident cells and fibroblasts (F) to restore tissue integrity.

expression and/or connectivity are altered in a number of inflammatory conditions in vitro and in vivo [21,22]. In vitro, inhibition of GJIC by proinflammatory mediators, such as lipopolysaccharide (LPS), tumor necrosis factor (TNF)-α, interleukin (IL)-1α, or IL-1β has been documented in endothelial cells [26,27], vascular smooth muscle cells [28], Schwann cells [29], astrocytes [30], and immortalized mouse hepatocytes [31]. Interestingly, the uncoupling of cultured rat hepatocytes induced by LPS required coculture with Kupffer cells, suggesting an indirect effect of the bacterial molecule [32]. Rapid closure of myoendothelial junctions was also reported in co-cultures of human umbilical vein endothelial and smooth muscle cells that were exposed to TNF-α [33]. Most of these rapid changes in GJIC might be explained by actions of the proinflammatory mediators on the gating and permeability properties of the gap junction channels involved. Indeed, phosphorylation at intracellular parts of connexins is known to alter the gating and permeability of the gap junction channels. For example, it was reported that the tyrosine kinase c-Src might mediate the rapid decrease in GJIC by TNF-α in human airway epithelial cell lines by phosphorylation of Cx43 channels [34]. Very recently, p38 stress-activated protein kinases were proposed to control gap junction closure in astrocytes by effecting protein kinase C, a molecule downstream in the p38 signaling cascade [35]. In addition, proinflammatory mediators may affect connexin gene expression. Thus, TNF-α induced the down-regulation of Cx43 expression in the rat heart by inhibition of the Cx43 promoter activity [36]. Although the precise mechanism remains to be elucidated, an AP-1 site in its promoter is likely involved in the cytokine regulation of Cx43 [37]. Similarly, circulating cytokines might be responsible for the reduction in connexin expression and GJIC along with gap junction redistribution observed in advanced stages of heart disease, as for example after ischemia and during heart failure [38–41].

In addition to their actions on gene expression, cytokines might affect the stability of connexin mRNAs or protein degradation. Indeed, the half-life of Cx32, which is the main connexin expressed in the liver, is reduced by LPS or after ischemia/reperfusion in mice and rats [42–45]. In the latter case, the decrease in Cx32 expression is likely to be the result of both accelerated degradation of Cx32 mRNA and disappearance of the connexin from the hepatocyte cell surface. Similarly, hepatoxicity induced by carbon tetrachloride (CCl_4), which is thought to activate the inflammatory cascade, and common bile duct ligation-induced cholestasis reduce the expression of Cx32 and Cx26 [46,47]. So far, it is not known whether these effects on connexins are consequences of regulation at the mRNA or protein levels. Possibly, the shortening of the poly(A) tail may affect the stability of Cx32 mRNA during inflammation [48]. In contrast to these findings, Temme et al. [45] showed in immortalized mouse hepatocytes that decreased expression of Cx32 in response to cytokine-mediated acute-phase response (IL-1, IL-6, TNF-α) was not associated with changes in the protein mRNA level. Finally, internal ribosome entry sites (IRES) have been identified in Cx32 and Cx43 mRNA that have been implicated in translational control of connexin expression [49,50]. Whether IRES are also implicated in cytokine-induced alterations in connexin expression remains to be investigated.

Although these observations point to gap junctions as important targets of inflammatory cytokines released after injury, there are few studies addressing whether a change in GJIC contributes to the severity of inflamed tissue or is a secondary consequence of tissue damage. In a spontaneous murine model of autoimmune thyroid disease, chronic inflammation reduced connexin (Cx26, Cx32 and Cx43) expression. Interestingly, communication-deficient diseased thyrocytes persisted in primary cultures, suggesting that the continued presence of inflammatory mediators is not required to sustain this difference [51]. More recently, Temme et al. [45] took advantage of the generation of Cx32-deficient mice to determine whether loss of this connexin gene effected LPS-induced inflammation. However, no difference between Cx32-deficient and wild-type liver acute-phase transcripts was detected. In the same animal model, the contribution of GJIC in cell response to injury was also analyzed during experimentally induced acute pancreatitis by injection of the pancreatic secretagogue cerulein. Interestingly, deficiency of Cx32 converted a mild reversible form of acute pancreatitis into a severe disease with increased necrosis, edema, and inflammation of the exocrine pancreas [52]. Importantly, Cx32-deficient mice exhibited decreased sensitivity of acinar cells to apoptotic stimuli. Proinflammatory cytokines are thought to exacerbate cell damage but also to play key roles in tissue repair by stimulating apoptosis and cell proliferation [53]. In the exocrine pancreas, apoptotic acinar cells failed to communicate with neighboring cells (M.C., unpublished data). Thus, GJIC may be important for the cells to defend themselves against inflammatory stimuli, for example by responding appropriately to apoptotic stimuli.

Gap junction channels are rapidly gated. Thus, modulation of GJIC may provide an early mechanism of cell defense against tissue-perturbed integrity and homeostasis, whether related to environmental or to genetic disease processes. In this context, it is worth to note that forced expression of connexins in astrocytes protects against cell injury induced by a variety of cell stress [54], whereas focal brain ischemia evoked in mice lacking connexin43 in astrocytes is associated with increased apoptosis and inflammation [55]. Hence, it is interesting to note that gating of gap junction channels between epithelial cells is defective in cystic fibrosis, a chronic infectious and inflammatory disease caused by mutations of the *CFTR* gene [56–58].

3. Tissue immune response

After injury or infections, the local production of cytokines and chemokines rapidly stimulates leukocyte extravasations and migration into tissues (Fig. 1). This process involves cell–cell contacts via adhesion molecules between leukocytes and endothelial cells during extravasations, as well as between leukocytes and target cells at sites of inflammation. The first blood cell members to appear after injury or infections are polymorphomononuclear cells (PMNs) of which the most abundant are neutrophils. There is circumstantial evidence that gap junctions may also be established between inflammatory cells during the course of inflammation [21]. It is now increasingly recognized that nonactivated leukocytes freshly extracted from peripheral blood do not express connexins [24,59]. Stimulation with LPS of peripheral hamster blood leukocytes, however, associates with the positive immunodetection of Cx43 [59]. In human, LPS induced the aggregation of PMNs, an event that did coincide with the translocation of Cx43 to the cell membrane. Microinjection of Lucifer Yellow in these aggregates did not result in dye coupling unless leukocytes were treated with conditioned medium from activated endothelial cells [60]. These observations suggest that soluble factors secreted by endothelial cells may be involved in the induction of PMNs to form gap junctions. In this context, we have evaluated GJIC in aggregated PMNs recovered from bronchoalveolar lavage (BAL) of mice that received intra-tracheal instillation of LPS. Under these conditions, PMNs, which were massively recruited from the blood stream and crossed the endothelial and airway epithelial barriers, did not exhibit cell coupling as evaluated by microinjection of Lucifer Yellow and neurobiotin (M.C., unpublished data). Clearly, additional experiments performed under conditions close to in vivo inflammatory situations are necessary to unequivocally address the question of gap junction formation between activated PMNs.

Second in line of defense against invading microorganisms and response to injuries are the macrophages, either already resident in the tissue or recruited as monocytes from the peripheral blood. Similar to the PMNs, nonactivated monocytes freshly extracted from peripheral blood do not express connexins [61]. Resident macrophages isolated from BAL of control mice also showed no intercellular diffusion of Lucifer Yellow or neurobiotin (M.C., unpublished data). As for PMNs, conditioned medium from endothelial cell cultures evoked translocation of connexins from the cytoplasmic compartment to the cell membrane and stimulate dye coupling in rat microglia [62]. Consistent with these findings, Cx43 has been detected in Kupffer cells in liver [32], peritoneal macrophages [59,63], kidney macrophages in inflammatory renal disease [64], microglia in brain [65], macrophage foam cells in atherosclerotic lesions [61,66], as well as in murine [63,67,68] and human monocytic/macrophage cell lines (Table 1). There is little evidence, however, for dye or electrical coupling between macrophages [69]. In the MH-S murine macrophage cell line treated or not with proinflammatory mediators, including granulocyte-macrophage colony stimulating factor (GM-CSF), interferon (IFN)-γ, and TNF-α, we could observe the diffusion of neurobiotin, but not Lucifer Yellow, in only 2 out of 15 cell clusters (Fig. 2). Of note, Cx43 could not be detected by Western blot in this mouse macrophage cell line (Fig. 2, Table 1), suggesting very low level of Cx43 expression or the involvement of a yet not identified connexin. Interestingly, Cx37 was detected in mouse and

Table 1
Connexin expression and GJIC in primary monocytes/macrophages and in monocyte/macrophage cell lines

Monocyte/macrophage	Connexin	Lucifer Yellow coupling	Reference
Human circulating	no Cx43	ND	[61]
Human circulating activated	Cx43	yes	[74]
Human macrophage foam cell	Cx37, Cx43	ND	[61,66]
Human kidney macrophage	Cx43	ND	[64]
Hamster circulating	no Cx43	no	[59]
Hamster circulating activated	Cx43	ND	[59]
Hamster peritoneal macrophage	Cx43	ND	[59]
Mouse peritoneal macrophage	Cx43	no	[63]
Mouse peritoneal macrophage	ND	no	unpublished data
Mouse peritoneal macrophage activated	Cx43	no	[63]
Mouse alveolar macrophage	ND	no	unpublished data
Mouse macrophage foam cell	Cx37, Cx43	ND	[24,66]
Mouse Langerhans cell	Cx43	ND	[21]
Rat microglia	Cx43	minimal	[62]
Rat microglia activated	Cx43	yes	[62]
Rat Kupffer cell	Cx43	ND	[31]
J774 (murine macrophage)	Cx43	yes	[63,67]
P388D1 (murine macrophage)	Cx43	yes	[68]
MH-S (murine monocyte)	no Cx43	no	unpublished data
THP-1 (human monocyte)	Cx43	no	unpublished data

ND: not determined. The absence of detection of specific connexins is indicated when determined.

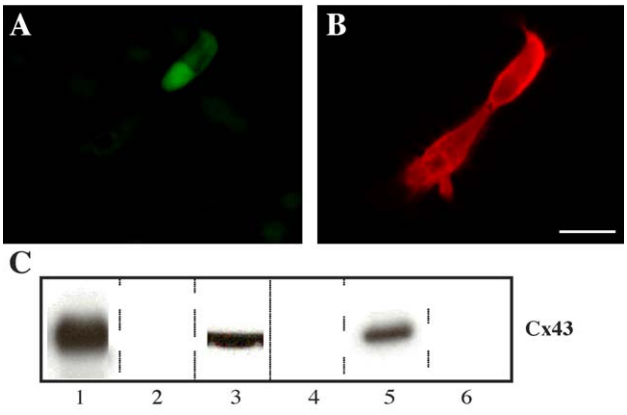

Fig. 2. GJIC and Cx43 expression in leukocytes. Co-microinjection of Lucifer Yellow (A) and neurobiotin (B) in a MH-S cell pair revealed cell–cell diffusion of the second tracer only. Bar represents 10 μm. Cx43 expression was detected by Western blots (C) in a control Cx43-expressing airway cell line (lane 1), in THP-1 cells (lane 3), and in a mixed population of human lymphocytes/monocytes isolated from buffy coats (lane 5). Cx43 was absent in the SKHep1 Cx43-deficient cell line (lane 2), in the MH-S cell line (lane 4) and in human neutrophils isolated from buffy coats (lane 6).

human macrophage foam cells (Fig. 3) [66]. This connexin is known to better permeate neurobiotin than the higher molecular weight dye Lucifer Yellow.

Upon repeated exposure to the same antigen (chronic infection and/or inflammation), the specific immune response enhances the function of innate immunity. Thus, a foreign molecule is internalized by phagocytosis and processed by antigen-presenting cells that migrate to secondary lymphoid organs where they interact with T helper cells. T helper cells either react directly with the antigen-bearing target cells (cytotoxic T cells) or they interact with B cells, which then differentiate and secrete specific immunoglobulins (humoral immunity). The possible role of gap junctions in the immunological synapse has been explored by Oviedo-Orta and Evans who first reported that circulating human T, B, and natural killer cells express Cx43 only, whereas human tonsil-derived T and B cells express Cx40 and Cx43 [70]. In mixed lymphocyte cultures, they observed that blocking GJIC markedly reduced the secretion of immunoglobulins and IL-10, suggesting that GJIC is an important component of the mechanisms underlying metabolic cooperation in the specific immune system [71]. Blocking of GJIC was achieved by either 18-α-glycyrrhetinic acid (αGA) or by synthetic peptides that show specific sequence overlap with the second extracellular loop of Cx40 or Cx43, thus preventing docking of their respective hemichannels. Although additional studies are needed to address the relevance of these observations, it has been proposed that gap junction expression and function might participate to the immune response, possibly by regulating leukocyte activation and recruitment [72,73].

Several laboratories have tested the possibility that gap junctions are involved in leukocyte transmigration in vitro. Thus, transmigration of leukocytes across an endothelial cell monolayer is altered in the presence of connexin-mimetic peptides or gap junction channel blockers [72–74]. In these studies, it has been shown that inhibition of GJIC increased transendothelial migration of neutrophils, decreased that of monocytes but had only modest effects on lymphocytes. We have also explored the transmigration of human leukocytes across monolayers of epithelial cells expressing different connexin types. In contrast to earlier studies, the trans-migration of PMNs across a monolayer of human CF airway cells expressing Cx43, treated or not with TNF-α, was not affected by adding uncoupling concentrations of αGA to the experimental medium (Fig. 4).

We further studied whether connexin-specific cell–cell signaling is involved in the transmigration of monocytes. To this end, the transmigration of THP-1 cells, a monocytic human cell line expressing Cx43 (Fig. 2), was evaluated in response to monocyte chemoattractant protein 1 (MCP-1). Transmigration was performed across monolayers of TNF-α-activated epithelial cells stably expressing one of the vascular connexins, namely Cx37, Cx40, or Cx43. The rate of THP-1 transmigration was not different across Cx37-, Cx40-, or Cx43-expressing cells but slower to that of parental communication-incompetent cells (J.P.D. and B.K., unpublished data). Again, αGA did not significantly effect monocyte transmigration across connexin-transfected cells in this assay (Fig. 4D). These conflicting reports may reflect variability between laboratories in the in vitro cell systems used, including the choice of epithelial/endothelial cell lines

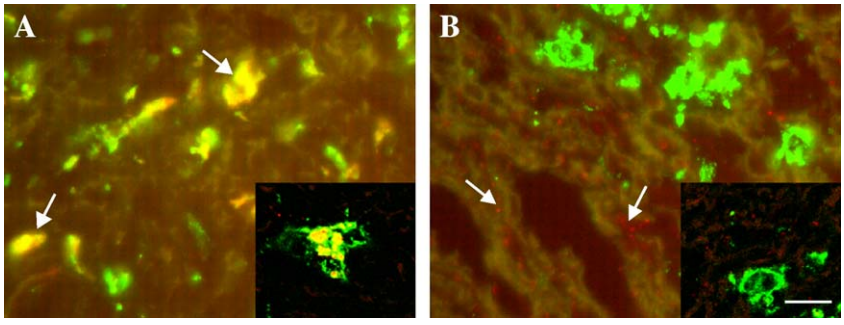

Fig. 3. Cx37 and Cx43 expression in human atherosclerotic plaques. Frozen sections (5 μm) of an advanced atheroma in the human carotid artery were double immunostained with antibodies against HAM-56 and against Cx37 (A) or Cx43 (B) (Texas Red detection). Sections were photographed using conventional fluorescence microscope. Inserts show images obtained by fluorescence confocal microscopy. Note that the macrophage marker HAM-56 (FITC detection) is co-localized with Cx37 (Texas Red detection and arrowheads in A), but not with Cx43 (arrowheads in B). Bar represents 50 μm.

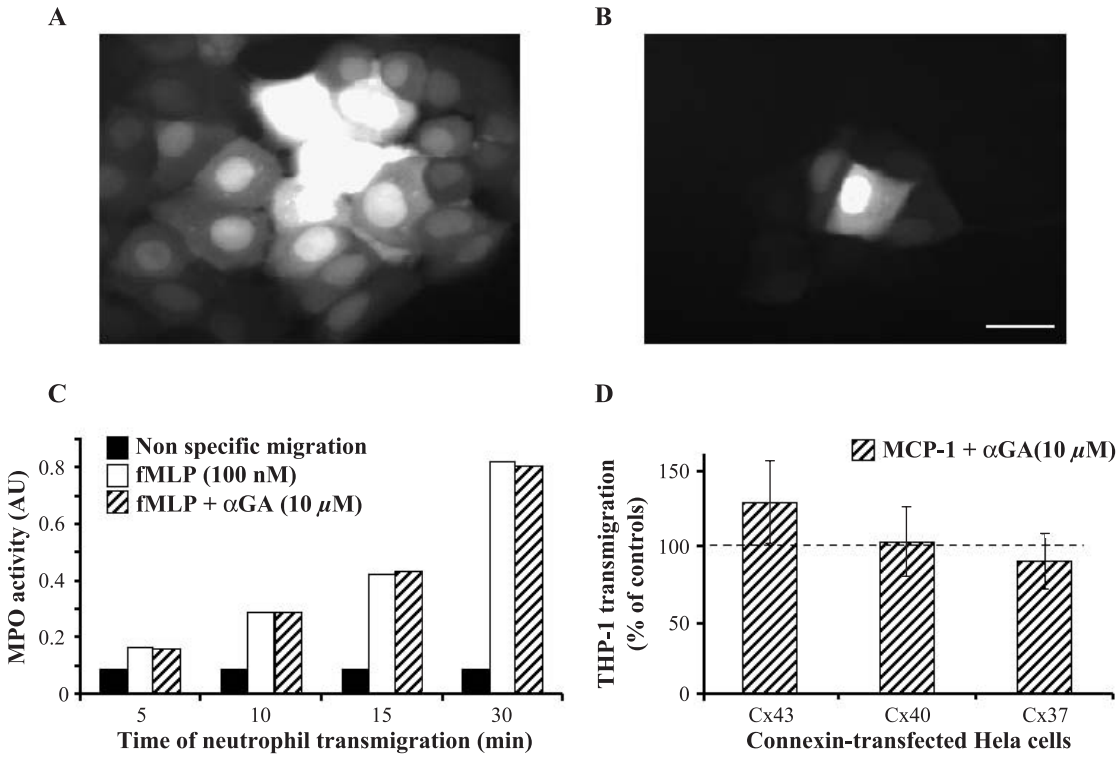

Fig. 4. Effect of αGA on the transmigration of leukocytes across a cell monolayer. (A, B) Uncoupling concentration of αGA was used, as revealed by the marked decrease of Lucifer Yellow diffusion (B) in contrast to normal dye coupling extent in a Cx43-expressing human CF15 airway cell line (A). Bar represents 10 μm. (C) Human neutrophils isolated from buffy coats were subjected to transmigration across CF15 cells in response to the chemotactic factor *n*-formyl methionyl leucyl phenylalanine (fMLP) and in the absence or presence of αGA. No change in the myeloperoxydase (MPO) activity of cells that transmigrated was observed with time, indicating that αGA did not affect neutrophil chemotactism. Each value is mean of four separate measurements in two experiments. The lack of effect of αGA in neutrophil transmigration was confirmed in additional experiments at 30 min (n=5), whatever the direction of cell migration (apical to basal or basal to apical) or the presence of TNF-α. (D) The transmigration of THP-1 cells in response to MCP-1 was performed across monolayers of TNF-α-activated epithelial cells stably expressing one of the vascular connexins. The rate of THP-1 transmigration across Cx37-, Cx40-, and Cx43-expressing cells was measured in the absence or presence of αGA. In all cell lines, αGA did not affect the rate of THP-1 transmigration. For each cell line, data were normalized to their respective controls in the absence of αGA.

and methods to isolate and activate leukocytes. Clearly, it will require additional studies in more physiological settings to unambiguously address whether connexin-associated functions are involved in leukocyte transmigration.

Although in vitro data showed apparent conflicting results, recent studies in LDL receptor-deficient ($LDLR^{-/-}$) mice, a mouse model for atherosclerosis, shed new light on the involvement of connexin in leukocyte recruitment to inflammatory sites. Initiation of atherosclerosis, a chronic inflammatory disease, involves complex patterns of interaction between leukocytes and the cells of the arterial wall, in which adhesion molecules, cytokines, and chemokines are known to play a critical role. During atherogenesis, induction of Cx43 expression has been observed in the dysfunctional endothelium at the shoulder of atherosclerotic lesions [66]. Interestingly, not only the progression of atherosclerosis was reduced by 50%, but atherosclerotic lesions also contained significantly less inflammatory cells in $LDLR^{-/-}$ mice intercrossed with mice heterozygous for a Cx43 null mutation ($Cx43^{+/-}$ $LDLR^{-/-}$ mice) [75]. In these mice, the expression of Cx43 is decreased by half in the aorta [76]. Although the underlying mechanisms remain to be determined, these results provide in vivo evidence for a key role of Cx43 in the development of an inflammatory disease. In the same line, it is worth mentioning that application of Cx43 antisense to wounded skin in mice reduced inflammatory parameters, including leukocyte immigration [77]. The challenge remains to identify whether (and which) signals are being exchanged through gap junctions between leukocytes and target cells during various phases of the inflammatory response. In this context, hemi-gap junction channels (connexons) at the plasma membrane may be more than precursors of gap junctions by allowing the transport in and out of the cells of signaling molecules [78,79]. Thus, the release of ATP through Cx26 hemichannels expressed in an epithelial cell line has been involved in invasion and dissemination of *Shigella flexneri*, a pathogen that causes intense inflammation and destruction of the colonic mucosa [80].

4. Tissue repair

The production of cytokines and of a variety of growth factors by the damaged tissue also triggers the migration of resident cells and fibroblasts to restore tissue integrity. Remodeling of the connective tissue occurs at final stages of wound healing, a process that is tightly regulated by a balance between cell proliferation and programmed cell death. Disruption of these processes leads to delayed wound healing and excessive scarring that may take months to resolve in adults. In a number of disease states, the resolution of scarring is impaired, leading to tissue fibrosis [81]. Many experimental and clinical studies have demonstrated the mostly beneficial effects of exogenous growth factors, such as GM-CSF, platelet-derived growth factor (PDGF), fibroblast growth factor (FGF), epidermal growth factor (EGF), and transforming growth factor β (TGF-β) on the healing process. However, the roles played by endogenous growth factors have remained largely unclear [82]. Although the immigration of inflammatory cells was believed to coordinate the repair process as a major source of wound growth factor signals, recent provocative observations contradicted this view. Indeed, the repair of a skin wound was found identical to wild-type animals in PU.1-deficient mice, which are genetically incapable of raising an inflammatory response [83]. These data indicate that cytokines and growth factors from inflammatory cells may not be essential for repair but might be causal of fibrosis at sites of adult healing.

The regulation of Cx43 expression and GJIC by exogenously applied growth factors has been extensively studied in cultured cells. The outcome varies between positive and inverse relationships dependent on the growth factor and the type of membrane receptors employed as well as on the initial activation/phosphorylation state of the cells. For example, EGF and related ligands signaling through receptor tyrosine kinases, transiently reduces GJIC via effects on channel gating by phosphorylation of Cx43 channels [84,85]. Other growth factors like TGF-β and FGF enhanced GJIC by increasing gap junction plaque formation [86–88], an effect likely mediated via activation of extracellular signal-regulated kinase (ERK) in lens [89]. Very variable results were obtained on gap junctions in different wound healing models. For example, connexin expression and GJIC were found to decrease after wounding of the rabbit corneal epithelium until completion of wound closure, a time point associated with re-establishment of GJIC [90,91]. In these studies, actively migrating epithelial cells no longer exhibited GJIC in the wounded area whereas another report on rabbit corneas showed no alteration in connexin expression by migrating cells [92]. In rat carotid artery after denudation injury, alternate changes in the expression of the three-endothelial connexins (Cx37, Cx40, Cx43) have been associated with ongoing regeneration of the arterial endothelium [93]. These observations suggest for different intercellular communication requirements during the various phases of the healing process. Indeed, the necessity for proper connexin expression to coordinate sheet migration during endothelial repair has been demonstrated by applying dominant negative connexin inhibitors in an in vitro endothelial wounding model [94].

Increasing evidence indicates that changes in GJIC between keratinocytes may help to coordinate cell proliferative and migratory activities at the healing margin of the wounded skin. The skin is known to express up to nine connexins that functionally compartmentalize the epidermis [20,95–97]. The immediate response to wounding (hours) is a decreased expression of all connexins in the epidermis (Cx43, Cx26, Cx30, and Cx31.1) and dermis [97,98]. However, the profile of connexin expression changes thereafter in parallel with key events in the wound healing process. By correlating connexin expression with Ki67 immunolabeling, Coutinho et al. [98] concluded that Cx26 and Cx30 reappear in non-proliferative, migrating keratinocytes at the wound leading edge. In contrast, Cx43 correlated with cell proliferation, behind the wound leading edge, while C31.1 reappeared during terminal differentiation of keratinocytes in the granular layer at 7 days post-wounding [98]. In wounded mouse-tail skin, Kretz et al. [99] observed similar changes but found increased expression of Cx31 into basal and then suprabasal layers of the epidermis on day 3 post-wounding.

The importance of time-dependent regulation of gap junctions during wound healing and skin repair has been highlighted by two recent studies. Indeed, treatment of the wound with Cx43 antisense dramatically increased the rate in wound closure [77]. It is important to note that the effects of Cx43 antisense were transient so that the re-expression of Cx43 in later post-wounding stages was not affected. Thus, down-regulating Cx43 in basal layer cells of the epidermis may favor the transformation or dedifferentiation of keratinocytes into a migrating phenotype. Similar conclusions were drawn from wound healing experiments performed in mice with a skin-specific deletion of Cx43 [99]. Again, lack of Cx43 accelerated wound closure in the tail skin of these mice. In contrast to Qiu et al. [77], however, the authors suggested that decreased GJIC between epidermal cells might also be necessary for mobilization and proliferation of keratinocytes. Clearly, additional experiments will be needed to undercover the specific roles of GJIC during the different phases of wound repair in the epidermis, a tissue where appears to be an extensive functional redundancy between connexins [20,99].

The multiple effects that were reported for wound treatment with Cx43 antisense on decreased inflammation and faster scar resorbtion are of particular importance but difficult to interpret. Antisense reduced Cx43 expression in the epidermis and dermis. Possibly, the treatment may alter the production of pro-inflammatory signals by the keratinocytes. In addition, other cell types expressing Cx43 may also be affected by the antisense, for example by altering the recruitment of leukocytes or perturbing the progression of fibroblasts through the wound repair process [100]. Although the mechanisms by which Cx43 antisense reduced inflammation was not uncovered, these results provide evidence

that local modulation of the cellular inflammatory response at the site of wounding might be a beneficial therapeutic strategy for management of tissue repair. Such strategies may not only be important for scarless and fast tissue repair, but may also be effective in case of excessive tissue repair reaction as observed, for example, after balloon angioplasty in coronary arteries. Indeed, important changes in connexin expression have been observed in vascular smooth muscle cells during restenosis- or atherosclerosis-related neointima formation [66,101]. Thus, gap junctions may represent important pharmacological targets for modulation of the inflammatory response.

5. Concluding remarks

While the first descriptions of electrical coupling between activated lymphocytes have been reported in the 1970s [102–104], surprisingly little mechanistic insight in the role of gap junctions during the inflammatory response following injury and during the repair process has been obtained. Outstanding reports have since described alterations in connexin expression in diseased human tissues. However, most of these studies focused only on connexin expression in the original tissue at a given time point after injury but little or no attention was given to the series of events leading to repair. The increasing availability of connexin-specific antibodies allowed us now to causally link the observed alterations in connexin expression to the progression of disease. Studies using conditional knockout mice where connexins are temporally and spatially deleted, which are likely underway should shed some light on these issues. Perhaps the most challenging task for researchers will be to unravel the precise steps that are influenced by gap junctions during the cascade of events underlying the inflammatory and repair processes. This leaves new and exciting future work in this field of research.

Acknowledgements

This work was supported by grants from the Swiss National Science Foundation (3100-067777.02 and PPOOA-68883 to BRK, and 3100-067120.01 to MC), the French Association Vaincre la Mucoviscidose (to MC) and the Swiss University Conference (SUK) program ‘Heart Remodeling in Health and Disease’ (to BRK).

References

[1] D.A. Goodenough, J.A. Goliger, D.L. Paul, Connexins, connexons and intercellular communication, Annu. Rev. Biochem. 65 (1996) 475–502.
[2] D.C. Spray, Molecular physiology of gap junction channels, Clin. Exp. Pharmacol. Physiol. 23 (1996) 1038–1040.
[3] A.L. Harris, Emerging issues of connexin channels: biophysics fills the gap, Q. Rev. Biophys. 34 (2001) 325–472.
[4] W.H. Evans, P.E. Martin, Gap junctions: structure and function, Mol. Membr. Biol. 19 (2002) 121–136.
[5] J.C. Saéz, V.M. Berthoud, M.C. Branes, A.D. Martinez, E.C. Beyer, Plasma membrane channels formed by connexins: their regulation and functions, Physiol. Rev. 83 (2003) 1359–1400.
[6] V.M. Unger, N.M. Kumar, N.B. Gilula, M. Yeager, Three-dimensional structure of a recombinant gap junction membrane channel, Science 283 (1999) 1176–1180.
[7] G.A. Perkins, D.A. Goodenough, G.E. Sosinsky, Formation of the gap junction intercellular channel requires a 30 degree rotation for interdigitating two apposing connexons, J. Mol. Biol. 277 (1998) 171–177.
[8] K. Willecke, J. Eiberger, J. Degen, D. Eckardt, A. Romualdi, M. Güldenagel, U. Deutsch, G. Söhl, Structural and functional diversity of connexin genes in the mouse and human genome, Biol. Chem. 383 (2002) 725–737.
[9] R.B. Stagg, W.H. Fletcher, The hormone-induced regulation of contact-dependent cell–cell communication by phosphorylation, End. Rev. 11 (1990) 302–325.
[10] C. Elfgang, R. Eckert, H. Lichtenberg-Frate, A. Butterweck, O. Traub, R.A. Klein, D.F. Hulser, K. Willecke, Specific permeability and selective formation of gap junction channels in connexin-transfected HeLa cells, J. Cell Biol. 129 (1995) 805–817.
[11] H. Niessen, H. Harz, P. Bedner, K. Kramer, K. Willecke, Selective permeability of different connexin channels to the second messenger inositol 1,4,5-trisphosphate, J. Cell. Sci. 113 (2000) 1365–1372.
[12] M.A. Thomas, S. Huang, A. Cokoja, O. Riccio, O. Staub, S. Suter, M. Chanson, Interaction of connexins with protein partners in the control of channel turnover and gating, Biol. Cell 94 (2002) 445–456.
[13] F. Lecanda, P.M. Warlow, S. Sheikh, F. Furlan, T.H. Steinberg, R. Civitelli, Connexin43 deficiency causes delayed ossification, craniofacial abnormalities, and osteoblast dysfunction, J. Cell Biol. 151 (2000) 931–944.
[14] A. Plum, G. Hallas, T. Magin, F. Dombrowski, A. Hagendorff, B. Schumacher, C. Wolpert, J.-S. Kim, W.H. Lamers, M. Evert, P. Meda, O. Traub, K. Willecke, Unique and shared functions of different connexins in mice, Curr. Biol. 10 (2000) 1083–1091.
[15] T.W. White, Unique and redundant connexin contributions to lens development, Science 295 (2002) 319–320.
[16] J.P. Stains, F. Lecanda, J. Screen, D.A. Towler, R. Civitelli, Gap junctional communication modulates gene transcription by altering the recruitment of Sp1 and Sp3 to connexin-response elements in osteoblast promoters, J. Biol. Chem. 278 (2003) 24377–24387.
[17] T.W. White, D.L. Paul, Genetic diseases and gene knockouts reveal diverse connexin functions, Annu. Rev. Physiol. 61 (1999) 283–310.
[18] J. Lamartine, G. Munhoz Essenfelder, Z. Kibar, I. Lanneluc, E. Callouet, D. Laoudj, G. Lemaitre, C. Hand, S.J. Hayflick, J. Zonana, S. Antonorakis, U. Radhakrishna, D.P. Kelsell, A.L. Christianson, A. Pitaval, V. DerKaloustian, C. Fraser, C. Blancher-Bardon, Mutations in GJB6 cause hidrotic ectodermal dysplasia, Nat. Genet. 26 (2000) 142–144.
[19] R. Rabionet, N. Lopez-Bigas, M.L. Arbones, X. Estivill, Connexin mutations in hearing loss, dermatological and neurological disorders, Trends Mol. Med. 8 (2002) 205–212.
[20] G. Richard, Connexin gene pathology, Clin. Exp. Dermatol. 28 (2003) 397–409.
[21] J.C. Sáez, R. Araya, M. Branes, M. Concha, J. Contreras, E. Eugenin, A. Martinez, F. Palisson, M. Sepulveda, Gap junctions in inflammatory responses: connexins, regulation and possible functional roles, Curr. Top. Membr. 49 (2000) 555–579.
[22] A. De Maio, V.L. Vega, J.E. Contreras, Gap junctions, homeostasis, and injury, J. Cell. Physiol. 191 (2002) 269–282.
[23] E. Oviedo-Orta, W.H. Evans, Gap junctions and connexin-mediated communication in the immune system, Biochim. Biophys. Acta 1662 (2004) 102–112.

[24] C.W. Wong, T. Christen, B.R. Kwak, Connexins in leukocytes: shuttling messages? Cardiovasc. Res. 62 (2004) 357–367.
[25] T. Nakase, C.C.G. Naus, Gap junctions and neurological disorders of the central nervous systems, Biochim. Biophys. Acta 1662 (2004) 149–158.
[26] V.W. Hu, H.Q. Xie, Interleukin-1 alpha suppresses gap junction-mediated intercellular communication in human endothelial cells, Exp. Cell Res. 213 (1994) 218–223.
[27] H.V. van Rijen, M.J. van Kempen, S. Postma, H.J. Jongsma, Tumour necrosis factor alpha alters the expression of connexin43, connexin40, and connexin37 in human umbilical vein endothelial cells, Cytokine 10 (1998) 258–264.
[28] A. Mensink, L.H. de Haan, C.M. Lakemond, C.A. Koelman, J.H. Koeman, Inhibition of gap junctional intercellular communication between primary human smooth muscle cells by tumor necrosis factor alpha, Carcinogenesis 16 (1995) 2063–2067.
[29] K.J. Chandross, Nerve injury and inflammatory cytokines modulate gap junctions in the peripheral nervous system, Glia 24 (1998) 21–31.
[30] J.P. Bolanos, J.M. Medina, Induction of nitric oxide synthase inhibits gap junction permeability in cultured rat astrocytes, J. Neurochem. 66 (1996) 2091–2099.
[31] A. Temme, O. Traub, K. Willecke, Downregulation of connexin32 protein and gap-junctional intercellular communication by cytokine-mediated acute-phase response in immortalized mouse hepatocytes, Cell Tissue Res. 294 (1998) 345–350.
[32] H.E. Gonzalez, E.A. Eugenin, G. Garces, N. Solis, M. Pizarro, L. Accatino, J.C. Sáez, Regulation of hepatic connexins in cholestasis: possible involvement of Kuppfer cells and inflammatory mediators, Am. J. Physiol.: Gasterointest. Liver Physiol. 282 (2002) G991–G1001.
[33] J. Hu, I.A. Cotgreave, Differential regulation of gap junctions by proinflammatory mediators in vitro, J. Clin. Invest. 99 (1997) 2312–2316.
[34] S. Huang, T. Dudez, I. Scerri, M.A. Thomas, B.N.G. Giepmans, S. Suter, M. Chanson, Defective activation of c-Src in cystic fibrosis airway epithelial cells results in loss of tumor necrosis factor-α-induced gap junction regulation, J. Biol. Chem. 278 (2003) 8326–8332.
[35] D. Zvalova, J. Cordier, M. Mesnil, M.P. Junier, H. Chneiweiss, p38/SAPK2 controls gap junction closure in astrocytes, Glia 46 (2004) 323–333.
[36] M. Fernandez-Cobo, C. Gingalewski, D. Drujan, A. De Maio, Downregulation of connexin 43 gene expression in rat heart during inflammation. The role of tumour necrosis factor, Cytokine 11 (1999) 216–224.
[37] M. Fernandez-Cobo, D. Stewart, D. Drujan, A. De Maio, Promoter activity of the rat connexin 43 gene in NRK cells, J. Cell. Biochem. 81 (2001) 514–522.
[38] N.S. Peters, C.R. Green, P.A. Poole-Wilson, N.J. Severs, Reduced content of connexin43 gap junctions in ventricular myocardium from hypertrophied and ischemic human hearts, Circulation 83 (1993) 864–875.
[39] M.A. Beardslee, G. Laing, E.C. Beyer, J.E. Saffitz, Rapid turnover of connexin43 in the adult heart, Circ. Res. 83 (1998) 629–635.
[40] D.L. Lerner, K.A. Yamada, R.B. Schuessler, J.E. Saffitz, Accelerated onset and increased incidence of ventricular arrhythmias induced by ischemia in Cx43-deficient mice, Circulation 101 (2000) 547–552.
[41] E. Dupont, T. Matsushita, R.A. Kaba, C. Vozzi, S.R. Coppen, N. Khan, R. Kaprielian, M.H. Yacoub, N.J. Severs, Altered connexin expression in human congestive heart failure, J. Mol. Cell. Cardiol. 33 (2001) 359–371.
[42] C. Gingalewski, K. Wang, M.G. Clemens, A. De Maio, Post-translational regulation of connexin32 expression in liver during acute inflammation, J. Cell. Physiol. 166 (1996) 461–467.
[43] C. Gingalewski, A. De Maio, Differential decrease in connexin32 expression in the ischemic and nonischemic regions of rat liver during ischemia/reperfusion, J. Cell. Physiol. 171 (1997) 20–27.
[44] A. De Maio, C. Gingalewski, N.G. Theodorakis, M.G. Clemens, Interruption of hepatic gap junctional communication in the rat during inflammation induced by bacterial lipopolysaccharide, Shock 14 (2000) 53–59.
[45] A. Temme, T. Ott, T. Haberberger, O. Traub, K. Willecke, Acute-phase response and circadian expression of connexin26 are not altered in connexin32-deficient mouse liver, Cell Tissue Res. 300 (2000) 111–117.
[46] J.C. Sáez, M.V.L. Bennett, D.C. Spray, Carbon tetrachloride at hepatotoxic levels blocks reversible gap junctions between rat hepatocytes, Science 236 (1987) 967–969.
[47] M.B. Fallon, M.H. Nathanson, A. Mennone, J.C. Sáez, A.D. Burghstahler, J.M. Anderson, Altered expression and function of hepatocyte gap junctions after common bile duct ligation in the rat, Am. J. Physiol., Cell Physiol. 268 (1995) C1186–C1194.
[48] N.G. Theodorakis, A. De Maio, Cx32 mRNA in rat liver: effects of inflammation on poly(A) tail distribution and mRNA degradation, Am. J. Physiol. 276 (1999) R1249–R1257.
[49] A. Schiavi, A. Hudder, R. Werner, Connexin43 mRNA contains a functional internal ribosome entry site, FEBS Lett. 464 (1999) 118–122.
[50] A. Hudder, R. Werner, Analysis of a Charcot-Marie-Tooth disease mutation reveals an essential internal ribosome entry site element in the connexin-32 gene, J. Biol. Chem. 275 (2000) 34586–34591.
[51] L.M. Green, J.P. Lazarus, M. LaBue, M.M. Shah, Reduced cell–cell communication in a spontaneous murine model of autoimmune thyroid disease, Endocrinology 136 (1995) 3611–3618.
[52] J.-L. Frossard, L. Rubbia-Brandt, M.A. Wallig, M. Benathan, T. Ott, P. Morel, A. Hadengue, S. Suter, K. Willecke, M. Chanson, Severe acute pancreatitis and reduced acinar cell apoptosis in the exocrine pancreas of mice deficient for the Cx32 gene, Gastroenterology 124 (2003) 481–493.
[53] M.I. Luster, P.P. Simonova, R.M. Gallucci, A. Bruccoleri, M.E. Blazka, B. Yucesoy, Role of inflammation in chemical-induced hepatotoxicity, Toxicol. Lett. 120 (2001) 317–321.
[54] J.H. Lin, J. Yang, S. Liu, T. Takano, X. Wang, Q. Gao, K. Willecke, M. Nedergaard, Connexin mediates gap junction-independent resistance to cellular injury, J. Neurosci. 23 (2003) 430–441.
[55] T. Nakase, G. Sohl, M. Theis, K. Willecke, C.C. Naus, Increased apoptosis and inflammation after focal brain ischemia in mice lacking connexin43 in astrocytes, Am. J. Pathol. 164 (2004) 2067–2075.
[56] M. Chanson, I. Scerri, S. Suter, Defective regulation of gap junctional coupling in cystic fibrosis pancreatic duct cells, J. Clin. Invest. 103 (1999) 1677–1684.
[57] M. Chanson, P.-Y. Berclaz, I. Scerri, T. Dudez, K. Wernke-Dollries, L. Pizurki, A. Pavirani, M.A. Fiedler, S. Suter, Regulation of gap junctional communication by a proinflammatory cytokine in cystic fibrosis transmembrane conductance regulator-expressing but not cystic fibrosis airway cells, Am. J. Pathol. 158 (2001) 1775–1784.
[58] S. Huang, L. Jornot, L. Wiszniewski, T. Rochat, S. Suter, J.S. Lacroix, M. Chanson, Src signaling links mediators of inflammation to Cx43 gap junction channels in primary and transformed CFTR-expressing airway cells, Cell Adhes. Commun. 10 (2003) 279–285.
[59] P.I. Jara, M.P. Boric, J.C. Saéz, Leukocytes express connexin 43 after activation with lipopolysaccharide and appear to form gap junctions with endothelial cells after ischemia–reperfusion, Proc. Natl. Acad. Sci. U. S. A. 18 (1995) 7011–7015.
[60] M.C. Branes, J.E. Contreras, J.C. Sáez, Activation of human polymorphonuclear cells induces formation of functional gap junctions and expression of connexins, Med. Sci. Monit. 8 (2002) BR313–BR323.
[61] D. Polacek, R. Lal, M.V. Volin, P.F. Davies, Gap junctional communication between vascular cells. Induction of connexin43 messenger RNA in macrophage foam cells of atherosclerotic lesions, Am. J. Pathol. 142 (1993) 593–606.

[62] E.A. Eugenin, D. Eckardt, M. Theis, K. Willecke, M.V. Bennett, J.C. Saéz, Microglia at brain stab wounds express connexin 43 and in vitro form functional gap junctions after treatment with interferon-gamma and tumor necrosis factor-alpha, Proc. Natl. Acad. Sci. U. S. A. 98 (2001) 4190–4195.
[63] L.A. Alves, R. Coutinho-Silva, P.M. Persechini, D.C. Spray, W. Savino, A.C. Campos de Carvalho, Are there functional gap junctions or junctional hemichannels in macrophages? Blood 88 (1996) 328–334.
[64] G.S. Hillis, L.A. Duthie, P.A. Brown, J.G. Simpson, A.M. MacLeod, N.E. Haites, Upregulation and co-localization of connexin43 and cellular adhesion molecules in inflammatory renal disease, J. Pathol. 182 (1997) 373–379.
[65] A.D. Martinez, E.A. Eugenin, M.C. Branes, M.V. Bennett, J.C. Saéz, Identification of second messengers that induce expression of functional gap junctions in microglia cultured from newborn rats, Brain Res. 943 (2002) 191–201.
[66] B.R. Kwak, F. Mulhaupt, N. Veillard, D.B. Gros, F. Mach, Altered pattern of vascular connexin expression in atherosclerotic plaques, Arterioscler. Thromb. Vasc. Biol. 22 (2002) 225–230.
[67] E.C. Beyer, T.H. Steinberg, Evidence that the gap junction protein connexin-43 is the ATP-induced pore of mouse macrophages, J. Biol. Chem. 266 (1991) 7971–7974.
[68] C.A. Martin, M.E. el-Sabban, L. Zhao, R. Burakoff, F.R. Homaidan, Adhesion and cytosolic dye transfer between macrophages and intestinal epithelial cells, Cell Adhes. Commun. 5 (1998) 83–95.
[69] J.A. Levy, R.M. Weiss, E.R. Dirksen, M.R. Rosen, Possible communication between murine macrophages oriented in linear chains in tissue culture, Exp. Cell Res. 103 (1976) 375–385.
[70] E. Oviedo-Orta, T. Hoy, W.H. Evans, Intercellular communication in the immune system: differential expression of connexin40 and 43, and perturbation of gap junction channel functions in peripheral blood and tonsil human lymphocyte subpopulations, Immunology 99 (2000) 578–590.
[71] E. Oviedo-Orta, P. Gasque, W.H. Evans, Immunoglobulin and cytokine expression in mixed lymphocyte cultures is reduced by disruption of gap junction intercellular communication, FASEB J. 15 (2001) 768–774.
[72] E. Oviedo-Orta, R.J. Errington, W.H. Evans, Gap junction intercellular communication during lymphocyte transendothelial migration, Cell Biol. Int. 26 (2002) 253–263.
[73] S. Zahler, A. Hoffmann, T. Gloe, U. Pohl, Gap junctional coupling between neutrophils and endothelial cells: a novel modulator of transendothelial migration, J. Leukoc. Biol. 73 (2003) 118–126.
[74] E.A. Eugenin, M.C. Branes, J.W. Berman, J.C. Saéz, TNF-alpha plus IFN-gamma induce connexin43 expression and formation of gap junctions between human monocytes/macrophages that enhance physiological responses, J. Immunol. 170 (2003) 1320–1328.
[75] B.R. Kwak, N. Veillard, G. Pelli, F. Mulhaupt, R.W. James, M. Chanson, F. Mach, Reduced connexin43 expression inhibits atherosclerotic lesion formation in low-density lipoprotein receptor-deficient mice, Circulation 107 (2003) 1033–1039.
[76] C.W. Wong, F. Burger, G. Pelli, F. Mach, B.R. Kwak, Dual benefit of reduced Cx43 on atherosclerosis in LDL receptor-deficient mice, Cell Adhes. Commun. 10 (2003) 395–400.
[77] C. Qiu, P. Coutinho, S. Frank, S. Franke, L.-Y. Law, P. Martin, C.R. Green, D.L. Becker, Targeting connexin43 expression accelerates the rate of wound repair, Curr. Biol. 13 (2003) 1697–1703.
[78] D.A. Goodenough, D.L. Paul, Beyond the gap: functions of unpaired connexon channels, Nat. Rev., Mol. Cell Biol. 4 (2003) 285–294.
[79] M.V. Bennett, J.E. Contreras, F.F. Bukauskas, J.C. Saéz, New roles for astrocytes: gap junction hemichannels have something to communicate, Trends Neurosci. 26 (2003) 610–617.
[80] G. Tran Van Nhieu, C. Blair, R. Bruzzone, M. Mesnil, P. Sansonetti, L. Combettes, Connexin-dependent inter-cellular communication increases invasion and dissemination of *Shigella* in epithelial cells, Nat. Cell Biol. 5 (2003) 720–726.
[81] S.E. Mutsaers, J.E. Bishop, G. McGrouter, G.J. Laurent, Mechanisms of tissue repair: from wound healing to fibrosis, Int. J. Cell Biol. 29 (1997) 5–17.
[82] S. Werner, R. Grose, Regulation of wound healing by growth factors and cytokines, Physiol. Rev. 83 (2003) 835–870.
[83] P. Martin, D. D'Souza, J. Martin, R. Grose, L. Cooper, R. Maki, S.R. McKercher, Wound healing in the PU.1 null mouse-Tissue repair is not dependent on inflammatory cells, Curr. Biol. 13 (2003) 1122–1128.
[84] M.Z. Hossain, A.L. Boynton, Regulation of Cx43 gap junctions: the gatekeeper and the password, Science's stke 2000 (2000) PE1.
[85] P.D. Lampe, A.F. Lau, Regulation of gap junctions by phosphorylation of connexins, Arch. Biochem. Biophys. 384 (2000) 205–215.
[86] M.S. Pepper, R. Montesano, A. el Aoumari, D. Gros, L. Orci, P. Meda, Coupling and connexin 43 expression in microvascular and large vessel endothelial cells, Am. J. Physiol. 262 (1992) C1246–C1257.
[87] D.F. Gibson, M.Z. Hossain, G.S. Goldberg, P. Acevedo, J.S. Bertram, The mitogenic effects of transforming growth factors beta 1 and beta 2 in C3H/10T1/2 cells occur in the presence of enhanced gap junctional communication, Cell Growth Differ. 5 (1994) 687–696.
[88] D.M. Larson, M.J. Wrobleski, G.D. Sagar, E.M. Westphale, E.C. Beyer, Differential regulation of connexin43 and connexin37 in endothelial cells by cell density, growth, and TGF-beta1, Am. J. Physiol. 272 (1997) C405–C415.
[89] A.C. Le, L.S. Musil, A novel role for FGF and extracellular signal-regulated kinase in gap junction-mediated intercellular communication in the lens, J. Cell Biol. 154 (2001) 197–216.
[90] M. Matic, I.N. Petrov, T. Rosenfeld, J.M. Wolosin, Alterations in connexin expression and cell communication in healing corneal epithelium, Invest. Ophtalmol. Visual Sci. 38 (1997) 600–609.
[91] K. Suzuki, T. Tanaka, M. Enoki, T. Nishida, Coordinated rassembly of the basement membrane and junctional proteins during corneal epithelial wound healing, Invest. Ophtalmol. Visual Sci. 41 (2000) 2495–2500.
[92] I. Ratkay-Traub, B. Hopp, Z. Dor, L. Dux, D.L. Becker, T. Krenacs, Regeneration of rabbit cornea following excimer laser photorefractive keratectomy: a study on gap junctions, epithelial junctions and epidermal growth factor receptor expression in correlation with cell proliferation, Exp. Eye Res. 73 (2001) 291–302.
[93] H.I. Yeh, Y.J. Lai, H.M. Chang, Y.S. Ko, N.J. Severs, C.H. Tsai, Multiple connexin expression in regenerating arterial endothelial gap junctions, Arterioscler. Thromb. Vasc. Biol. 20 (2000) 1753–1762.
[94] B.R. Kwak, M.S. Pepper, D.B. Gros, P. Meda, Inhibition of endothelial wound repair by dominant negative connexin inhibitors, Mol. Biol. Cell 12 (2001) 831–845.
[95] D. Salomon, J.-H. Saurat, P. Meda, Cell-to-cell communication within intact human skin, J. Clin. Invest. 82 (1988) 248–254.
[96] B. Risek, F.G. Klier, N. Gilula, Multiple gap junction genes are utilized during rat skin and hair development, Development 116 (1992) 639–651.
[97] J.A. Goliger, D.L. Paul, Wounding alters epidermal connexin expression and gap junction-mediated intercellular communication, Mol. Biol. Cell 6 (1995) 1491–1501.
[98] P. Coutinho, C. Qiu, S. Frank, K. Tamber, D. Becker, Dynamic changes in connexin expression correlate with key events in the wound healing process, Cell Biol. Int. 27 (2003) 525–541.
[99] M. Kretz, C. Euwens, S. Hombach, D. Eckardt, B. Teubner, O. Traub, K. Willecke, T. Ott, Altered connexin expression and wound

healing in the epidermis of connexin-deficient mice, J. Cell. Sci. 116 (2003) 3443–3452.
[100] H.P. Ehrlich, T. Diez, Role of gap junctional intercellular communications in wound repair, Wound Repair Regen. 11 (2003) 481–489.
[101] H.I. Yeh, F. Lupu, E. Dupont, N.J. Severs, Upregulation of connexin43 gap junctions between smooth muscle cells after balloon catheter injury in the rat carotid artery, Arterioscler. Thromb. Vasc. Biol. 17 (1997) 3174–3184.
[102] D.F. Hulser, J.H. Peters, Intercellular communication in phytohemagglutinin-induced lymphocyte agglutinates, Eur. J. Immunol. 1 (1971) 494–495.
[103] G.M. Oliveira-Castro, M.A. Barcinski, S. Cukierman, Intercellular communication in stimulated human lymphocytes, J. Immunol. 11 (1973) 1616–1619.
[104] J.A. Levy, R.M. Weiss, E.R. Dirksen, M.R. Rosen, Possible communication between murine macrophages oriented in linear chains in tissue culture, Exp. Cell Res. 103 (1976) 375–385.