REVIEW ARTICLE

The Role of Neuromediators and Innervation in Cutaneous Wound Healing

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The skin is densely innervated with an intricate network of cutaneous nerves, neuromediators and specific receptors which influence a variety of physiological and disease processes. There is emerging evidence that cutaneous innervation may play an important role in mediating wound healing. This review aims to comprehensively examine the evidence that signifies the role of innervation during the overlapping stages of cutaneous wound healing. Numerous neuropeptides that are secreted by the sensory and autonomic nerve fibres play an essential part during the distinct phases of wound healing. Delayed wound healing in diabetes and fetal cutaneous regeneration following wounding further highlights the pivotal role skin innervation and its associated neuromediators play in wound healing. Understanding the mechanisms via which cutaneous innervation modulates wound healing in both the adult and fetus will provide opportunities to develop therapeutic devices which could manipulate skin innervation to aid wound healing.
Key words: wound healing; skin innervation; neuromediators; neuropeptides; fetal wound healing.

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Wound healing involves a set of highly regulated overlapping mechanisms that utilise cellular, molecular and chemical components. It begins at the moment of injury and continues for months or years to produce a healed wound that is similar to uninjured skin in terms of function and appearance. The central nervous system-skin interactions involve a number of neuromediators, cytokines, hormones and other effector molecules (1). Therefore stimuli received at the skin interface can influence the nervous system at both a local and central level; the brain can modulate skin function in physiological and pathophysiological states and the skin can alter the pharmacology of the central nervous system by releasing large amounts of neuropeptides (2).

The aim of this review is to provide a detailed update on normal skin innervation and its role in the different phases of wound healing. Furthermore, it describes the effect of cutaneous denervation on wounds in animal models and human clinical observations. Finally, the role of innervation in fetal wound healing which is regenerative rather than reparative is discussed.

SKIN INNERVATION

Skin contains a complex network of sensory nerve fibres (Fig. 1). The innervated skin is a vital barrier with direct contact to the central nervous system. These nerves are crucial in influencing physiological and pathophysiological cutaneous functions (3). Mechanoreceptors, thermoreceptors and nociceptors are found in the epidermis and dermis. Mechanoreceptors in the epidermis are Merkel discs and free nerve endings. Mechanoreceptors in the dermis are Ruffini, Meissner, Pacinian corpuscles and free nerve endings (4, 5).

Two distinct groups of nerve fibres are found in the skin. Cutaneous sensory nerve fibres are in close contact with dermal blood vessels, mast cells, fibroblasts, keratinocytes and Langerhans cells in the epidermis (6–9). Cutaneous sensory nerves are classified according to diameter and speed of impulse as Aβ, Aδ and C nerve
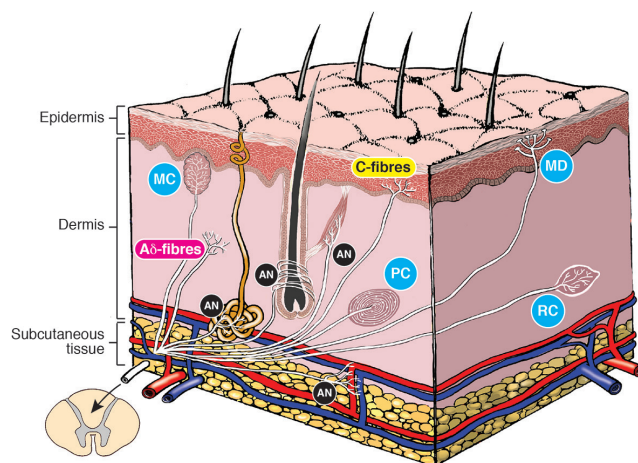

Fig. 1. Cross section outlining the distribution of the cutaneous nervous system in the skin. MC: Meissner's corpuscles; MD: Merkel disc; PC: Pacinian corpuscle; RC: Ruffini's corpuscle; AN: autonomic nerves.

fibres. Aβ fibres are fast and large whereas C fibres are slow and small. Aδ fibres constitute 80% of primary sensory nerves emerging from the dorsal root ganglia, whereas C fibres make up 20% of the primary nerves (10, 11). Aβ and Aδ are myelinated by accompanying Schwann cells. These sensory nerves, which extend throughout all layers of the skin (12), transfer signals from mechanoreceptors, thermoreceptors and nociceptors to their origin in the dorsal root ganglia. Mechanical stimuli are detected via mechanoreceptors associated with sensory corpuscles through Aβ fibres or with Aδ fibres; temperature via the thermoreceptors through Aδ and C fibres; and pain via nociceptors through Aδ and C fibres (3). From there, sensations like pain, burning and itching are forwarded to specific areas in the brain. The second group of nerves comprises the autonomic nerve fibres which constitute only a minority of cutaneous nerve fibres compared with sensory nerves. They are restricted to the dermis and are involved in regulating blood circulation, lymphatic function and the regulation of skin appendages (sweat glands, apocrine glands and hair follicles) (13, 14).

Sensory nerves in the skin are able to release neuromediators such as neuropeptides which signal to the skin (15, 16). In un-stimulated nerves, neuromediators are barely detectable within the skin tissues. However on direct stimulation a significant increase of regulatory neuromediators can be detected *in vitro* and *in vivo* (3). Neuropeptides are a family of extracellular messengers, which act as neurotransmitters, hormones or paracrine factors (17). The majority consist of a group of small peptides that exert their effects by interacting with members of a superfamily of G protein-coupled receptors (3). Numerous neuropeptides are expressed and released from sensory as well as autonomic cutaneous nerves, including calcitonin gene related peptide (CGRP), substance P, neurokinin A, and vasoactive intestinal peptide (VIP) (13). In addition, cutaneous cells themselves such as keratinocytes, micro-vascular endothelial cells, merkel cells, fibroblasts or leukocytes are capable of releasing neuropeptides (18, 19). The presence of neuropeptides in free nerve fibre endings and the proximity of these endings to a variety of cells in the skin seem to associate the cutaneous nervous system not merely for its role in sensation but in other biological actions as well, namely wound healing (20).

WOUND HEALING PHASES AND ROLE OF NEUROMEDIATORS

Wound healing involves a range of processes that operate in a systematic and timely manner to repair the skin's integrity and function. Wound healing can be divided into 3 overlapping stages: inflammation, proliferation and remodelling. There is increasing evidence that cutaneous innervation may play an important role in mediating normal wound healing. Numerous neuropeptides that are secreted by the sensory and autonomic nerve fibres play an essential part during the distinct phases of wound healing (Table I). The role of skin innervation and neuropeptides in the different phases of wound healing are discussed below.

Table I. *The role of neuromediators in cutaneous wound healing*

Neuromediator	Wound healing action
BDNF	Re-epithelialisation/keratinocyte proliferation
	Wound contraction
	Nerve regeneration
CGRP	Vasodilatation
	Release of pro-inflammatory cytokines
	Re-epithelialisation/keratinocyte proliferation
	Granulation tissue formation/fibroblast proliferation
	Angiogenesis
	Wound contraction
	Collagen maturation and remodelling
CRH	Release of pro-inflammatory cytokines
	Angiogenesis
SP	Vasodilatation
	Polymorphonuclear cell infiltration
	Release of pro-inflammatory cytokines
	Re-epithelialisation/keratinocyte proliferation
	Granulation tissue formation/fibroblast proliferation
	Angiogenesis
	Collagen maturation and remodelling
	Nerve regeneration
NPY	Vasoconstriction
	Angiogenesis
Gal	Vasoconstriction
	Release of pro-inflammatory cytokines
	Re-epithelialisation/keratinocyte proliferation
	Nerve regeneration
GRP	Re-epithelialisation/keratinocyte proliferation
	Angiogenesis
NGF	Re-epithelialisation/keratinocyte proliferation
	Angiogenesis
	Wound contraction
	Fibroblast differentiation into myofibroblasts
	Collagen maturation and remodelling
	Nerve regeneration
NKA	Vasodilatation
	Polymorphonuclear cell infiltration
	Re-epithelialisation/keratinocyte proliferation
	Granulation tissue formation/fibroblast proliferation
	Angiogenesis
	Nerve regeneration
NT-3	Nerve regeneration
PACAP	Vasodilatation
	Polymorphonuclear cell infiltration
	Re-epithelialisation/keratinocyte proliferation
VIP	Vasodilatation
	Release of pro-inflammatory cytokines
	Re-epithelialisation/keratinocyte proliferation (-ve effect)
	Angiogenesis
	Collagen deposition
	Nerve regeneration

BDNF: brain-derived neurotrophic factor; CGRP: calcitonin gene related peptide; CRH: corticotropin-releasing hormone; SP: Substance P; NPY: Neuropeptide Y; Gal: Galanoin; GRP: gastrin releasing peptide; NGF: nerve growth factor; NKA: neurokinin A; NT-3: Neurotrophin-3; PACAP: pituitary adenylate cyclase activating peptide ; VIP: vasoactive intestinal peptide.

Inflammatory phase

After a cutaneous injury, haemostasis which lasts a couple of hours produces a fibrin plug. The aggregated platelets release pro-inflammatory mediators such as cytokines and growth factors. These chemotactic signals recruit neutrophils which are the principal cellular components in the activation of the inflammatory phase of wound healing (21). Neutrophils act as chemo-attractants for other cells that are involved in the inflammatory phase (22), including modulating the expression of macrophages (23). Macrophages enter the wound and support the ongoing process by phagocytosis of pathogens and cell debris (24, 25) as well as by the secretion of growth factors, chemokines and cytokines (26, 27). The inflammatory phase can last between hours and days. Numerous neuropeptides released from the cutaneous innervation have been shown to activate vital processes during the inflammatory phase of wound healing (Fig. 2). Substance P (SP) appears to play a major role in the inflammatory phase; however other neuropeptides have also been identified and are reviewed.

SP is produced in the dorsal root ganglion of the spinal cord and distributed to the dorsal horn of the spinal cord and the nerve endings of the sensory neurons in the dermis and epidermis (17, 28). SP is also detected on human keratinocytes, endothelial cells and fibroblasts (29). The actions of SP are regulated through a neurokinin G-protein coupled receptor, NK-1R, which is expressed in neurons and peripheral tissues (30). SP acts on the vasculature, cutaneous epithelium and connective tissue. Nitric oxide is essential for adequate wound healing (31). SP stimulates vasodilatation and micro-vascular permeability through increasing nitric oxide release and through direct effects on endothelial cells (32). SP up-regulates expression of adhesion molecules on endothelial cells, monocyte chemotaxis and inflammatory cell activity (33–35). SP also modulates
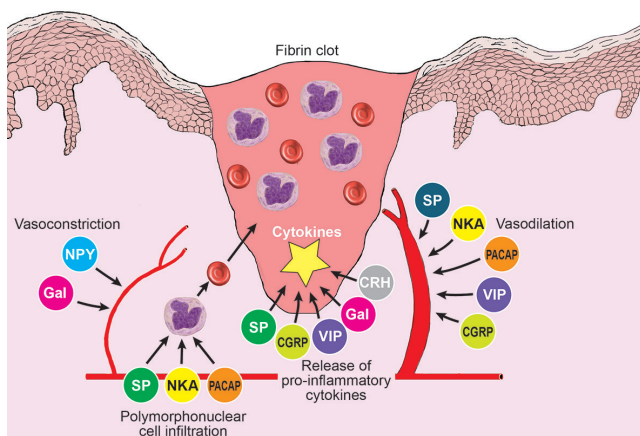
the synthesis and release of pro-inflammatory cytokines such as interleukins, transforming growth factor-α (TGF-α) and tumour necrosis factor-α; key components during the inflammatory phase of wound healing (36).

Neutral endopeptidase (NEP) is a zinc metalloprotease which competes with NK-1R and inhibits the actions of SP through enzymatic degradation (37). The interactions of SP and NEP are key components of inflammatory signalling in wound repair (20).

Neurokinin A is a bio-active tachykinin released into the skin after injury and activates, preferentially through neurokinin-2 receptor NK-2R, cutaneous target cells such as keratinocytes and dermal endothelial cells thereby modulating skin inflammation during wound healing (30). Immunohistochemistry studies have demonstrated corticotropin-releasing hormone (CRH) is found in sensory cutaneous nerves (38). CRH acts as a pro-inflammatory mediator and induces skin mast cell degranulation, thereby increasing vascular permeability and the release of pro-inflammatory cytokines (3). CRH has also been shown to enhance angiogenesis in the skin (39).

CGRP is a vasodilator and enhances plasma extravasation (40) and can stimulate angiogenesis. CGRP has been shown to increase the inflammatory response of other mediators such as SP (41). Activin is a member of the TGF-β superfamily which increases on wounding and has been shown to up-regulate CGRP expression in innervating sensory neurons (42), highlighting its regulatory role in wound healing. Nerve growth factor (NGF) has been shown to increase the release of CGRP from peripheral nerve terminals into peripheral tissue underlining its role as a modulator of the inflammatory phase of wound healing (43).

Kim & Pomeranz (44) showed that sympathetic nervous system stimulation accelerates wound healing at the dermal and epidermal levels through its proposed action on the process of neurogenic inflammation. Peripheral nerve fibres express α1-adrenoceptors and their expression increases after nerve injury possibly boosting neurogenic inflammation (45).

Proliferative phase

The early inflammatory phase is succeeded by a proliferative phase lasting several weeks. It is characterised by the invasion of macrophages and fibroblasts into the wounded region and formation of granulation tissue. Macrophages induce fibroblasts to produce an extracellular matrix (ECM) of type-3 collagen that provides a structural framework for endothelial cells, angiogenesis and wound contraction (Fig. 3).

SP has potent proliferative effects on fibroblasts, keratinocytes and endothelial cells by stimulating DNA synthesis (46, 47). It also stimulates angiogenesis through possible nitric oxide mediation (48, 49). SP

Fig. 2. Inflammatory phase. Overview outlining the role of neuromediators on the different wound healing actions during the inflammatory phase.
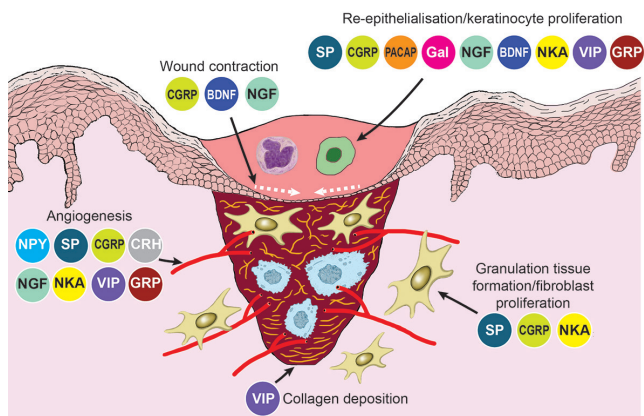
Fig. 3. Proliferative phase. Overview outlining the role of neuromediators on the different wound healing actions during the proliferative phase.

plays a crucial role in the granulation tissue remodelling process by promoting the proliferation and the migration of dermal fibroblasts (50) and by stimulating the expression of epidermal growth factor and its associated receptor (50, 51).

NGF is a polypeptide neurotrophin found in neurons of the central and peripheral nervous system as well as in a variety of cells including fibroblasts, epithelial cells, keratinocytes and immune cells (52). NGF has an essential role in differentiation, function and survival of sensory and autonomic nerves (53). NGF also has anti-inflammatory properties (54). NGF has been postulated to promote the proliferation of local immature cells in wounds, blood vessel formation and neurite overgrowth (55). NGF has been shown in animal studies and in a human case study to promote angiogenesis and epithelial healing (30, 55). Neurokinin A stimulates the release of NGF in the epidermis (56).

Other important neuropeptides identified to play a role in the proliferative phase include gastrin releasing peptide (GRP), CGRP, galanin, VIP and pituitary adenylate cyclase activating peptide (PACAP). GRP is widely distributed in the central and peripheral nervous system and although its implications in wound healing are not certain, some studies have shown it could promote migration, proliferation of keratinocytes and angiogenesis (57). CGRP stimulates the proliferation and migration of keratinocytes (58). Galanin is a peptide released from afferent nerves which signals through G-protein coupled receptors and has anti-proliferative effects in tissue (59). On the contrary an *in vitro* study identified galanin induced the up-regulation of NGF (60). VIP has been shown to act as a growth factor for proliferation of keratinocytes and as a modulator of their migration (61) as well as inducing histamine release by mast cells causing vasodilatation (62). VIP may be involved in the re-innervation of wounded tissue as it has been shown to promote sciatic nerve regeneration in rat sciatic nerve after transection (63). PACAP is found in sensory cutaneous nerves (64). It is a member of the

VIP peptide family and is a potent vasodilatator (65). It is postulated that C-fibres release PACAP in response to neuronal activation which in turn leads vasodilatation and extravasation. PACAP is involved in cutaneous inflammation by releasing histamine from mast cells and it also promotes human keratinocyte proliferation (66).

Activation and antagonism of the sympathetic nervous system via adrenoceptors during the proliferative phase of wound healing has been studied. A1-adrenoceptors influence growth cycles via mitogen activated protein kinase signalling pathways thus regulating cellular proliferation after injury (67). Activation of α1β-adrenoceptors and the β2-adrenoceptors mediates the proliferation and migration of fibroblasts (68). Whereas, β1/β2-adrenoceptor blockade accelerates human keratinocyte migration and re-epithelialisation in wound healing models (69). The production and remodelling of connective tissues in the skin are influenced by the balance of several essential cytokines, including TGF-β and insulin like growth factor-1 (IGF-1) (70). Studies have shown TGF-β stimulates overall ECM formation (71) and IGF-1 plays a regulatory role in enhancing proliferation of fibroblasts and modulates chemotaxis (72). Sympathetic stimulation through α-adrenoceptors elevates the production of TGF-β1 and IGF-1 in skin fibroblasts. These results imply that the sympathetic nervous system contributes to the modulation of cytokine secretion, ECM production and fibroblast migration in the skin (73). Neuropeptide Y (NPY) is widely distributed in the central and peripheral nervous system. In the skin the expression of NPY was detected in sympathetic nerve fibres in the deep and superficial dermis (74). NPY directly stimulates endothelial cell proliferation and migration, however its role as an angiogenic factor is debatable (75, 76).

Remodelling phase

The remodelling phase is characterised by proliferative cell apoptosis, ECM adjustment and organisation and replacement of type-3 with type-1 collagen (Fig. 4). The
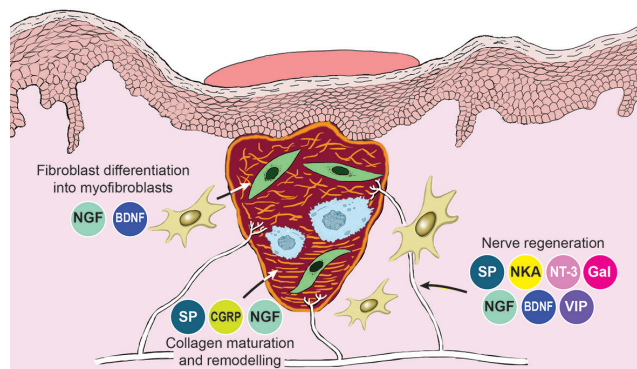

Fig. 4. Remodelling phase. Overview outlining the role of neuromediators on the different wound healing actions during the remodelling phase.

remodelling phase can last between weeks to years. Very little is known of the function of cutaneous innervation or neuropeptides in the remodelling phase.

Altun et al. (77) demonstrated significantly higher number of nerve fibres in normotrophic scars in comparison to hypertrophic scars suggesting a regulatory role for skin nerves in the remodelling phase of wound healing. Sensory nerve fibres regenerate into the repaired epidermis and dermis in response to neuropeptides (78). SP induces human dermal micro-vascular endothelial cells to produce NGF *in vitro* which in turn is required for nerve fibre regeneration following cutaneous injury (79). NGF has also been suggested to accelerate tissue remodelling (55). Neurotrophin-3 (NT-3) is a neurotrophic growth factor expressed by sensory and sympathetic nerves which is essential for growth, proliferation, and maintenance of nerves (80). Likewise brain-derived neurotrophic factor (BDNF) is required for the postnatal survival or functional maturation of sensory neurons. BDNF and their receptors are expressed by keratinocytes, fibroblasts and myofibroblasts, and promote their proliferation and differentiation (81, 82).

SP has been shown to influence the process of wound collagen degradation by increasing the matrix matalloproteinase-2 activity in fibroblasts (83). Fujiwara et al. (84) showed direct neuronal contact *in vitro* accelerates differentiation of fibroblasts into myofibroblasts which in turn secrete collagen fibres and induce wound contraction. Cheret et al. (85) showed SP, CGRP and VIP modulate matrix metalloproteinase activities and affect collagen-1 and collagen-3 productions during skin wound healing.

WOUND HEALING IN DENERVATED SKIN

Delayed wound healing in denervated skin further highlights the significance of cutaneous innervation in wound repair. Denervated skin is not simply associated with wounding but has a major role to play in the delayed healing process. This has been investigated experimentally in human and animal models and observed clinically.

Experimental denervation

Experimentally induced denervation supports the key role of skin innervation in wound healing. Vanilloid receptor-1 is expressed on sensory Aδ and C-fibres; and capsaicin is an agonist which causes denervation of these fibres. Capsaicin induced sensory denervation results in delayed re-epithelialisation, absence of vasodilatation and plasma protein extravasation indicating that cutaneous nerves are responsible for the initial two phases of wound healing (86, 87). Chemical sympathectomy with 6-hydroxydopamine delays wound healing by its effect on the inflammatory phase (88).

Denervated skin has been shown to exhibit delayed wound contraction and reduced micro-vascular response (89, 90). Fukai et al. (91) showed both wound contraction and epithelisation were delayed in denervated skin of mice by 17% and 25% respectively. Engin et al. (89) showed delayed wound contraction and loss of neuropeptide secretion from nerve endings in denervated tissue. The lack of neuropeptides in denervated skin may be the cause of deficiencies seen in the wound healing process. This hypothesis is enhanced by the finding that SP levels are decreased in denervated tissue and exogenous administration of SP promotes wound healing in mice (92–94). Likewise, capsaicin induced denervation lowers NGF transport (95) and differing levels of exogenous NGF accelerates wound closure, epithelisation and wound contraction (96, 97). Also, CGRP immune-reactive nerves are reduced in wounded tissue following denervation (92) and the role of CGRP in wound healing have been detailed above.

Diabetes mellitus

Diabetes is an important risk factor for the development and persistence of chronic wounds and all phases of wound healing are affected. Neuropathy is a possible cause of delayed wound healing in diabetes. There are fewer nerves in the epidermis and dermis in the skin of diabetic humans and mice (93). Animal studies have shown that diabetic mice have slower wound healing compared with controls and have delayed inflammatory cell migration (98). Neuropathy itself hinders the rate of cutaneous nerve regeneration (99) and Cheng et al. (100) showed that diabetic mice lack the capabilities to regenerate nerves. Neuropeptide expression and functions are affected in diabetes. Reduced SP and NGF positive nerve fibres are observed in diabetic patients and rats (101, 102); as well as reduced NGF and SP levels in the skin and serum (102, 103). This could also account for the decrease in angiogenesis seen in this sub-group of patients (104). NPY levels in diabetic skin are reduced (105). NPY binding to Y2 receptors is important in angiogenesis and as a result deletion of the Y2 receptor in diabetic mice results in the blockage of NPY induced angiogenesis and delays wound healing (75). NEP regulates the biologic action of SP. NEP levels are increased in diabetic wounds which may contribute to the deficient neuro-inflammatory signalling and wound healing in diabetics by decreasing the neuropeptide levels (106). The expression, release and action of CGRP is decreased in diabetes resulting in reduced CGRP mediated vasodilatation (107).

Paraplegic patients

The clinical observation that paraplegic and quadriplegic patients present with impaired wound closure below the level of the spinal cord lesion suggests the

participation of the nervous system in wound healing (108). Below the level of injury there is interruption of spinal vasomotor pathways resulting in a loss of tone in the vascular bed below the lesion and a state of generalised vasodilation (109). The denervated tissue is therefore unable to mount an inflammatory response (110) and the lack of tissue perfusion due to a chronic state of hypotension leads to the inability to deliver essential components to the wound (111, 112). This leads to lack of oxygen delivery to the wound which is vital for numerous steps of the wound healing cascade including angiogenesis, keratinocyte and fibroblast proliferation, collagen production and remodelling (113, 114). Further observations show wound healing is delayed in elderly humans (115). Khalil et al. (116) showed older rats heal substantially more slowly than those wounded at a younger age. All phases of wound healing are affected by age including delayed inflammatory infiltration into the wound, angiogenesis and collagen production and remodelling. A reduction in nerve endings as the skin ages could be a causative factor as to the delayed wound healing seen with ageing (117). Another human example is the delayed cutaneous wound healing that is apparent in those with leprosy induced neuropathy (118).

INFLUENCE OF PAIN ON WOUND HEALING

Cutaneous wounds are inevitably associated with pain. Common pain mediators released on wounding are SP, CGRP, nitric oxide and TNF-α. In acute wound healing, a short period of pain mediator release promotes the inflammatory phase and also enhances fibroblast collagen production, cellular migration and re-epithelialisation (119,120). However, in chronic pain conditions a resultant sustained release of these mediators creates an imbalance and may impede wound healing. SP is responsible for transmission of pain from sensory neurons to the central nervous system. The initial role of SP as a mediator of wound healing has been described above and the depleted levels of SP noted in diabetic patients may contribute to impaired cutaneous wound healing that is evident in this sub-group (121, 122). However prolonged pain and over-expression of SP could lead to a chronic inflammatory wound state (123). Similarly CGRP is proposed to contribute to pain transmission and has a key role to play in wound healing. However, as with SP, over-expression is detrimental to wound healing (124). TNF-α and nitric oxide are two important pain mediators which have crucial roles in wound healing. However, increased levels leads to chronic wound inflammation as seen in non healing wounds with impaired collagen production and anti-proliferative effects on fibroblasts (120, 123, 125). These findings are corroborated in a clinical study by McGuire et al. (126) who reported persistent post-surgery pain was an important predictor of healing time. They showed those that reported milder levels of pain post surgery experienced faster healing compared with those who reported more intense pain (126).

FETAL WOUND HEALING AND INNERVATION

Post natal cutaneous wound healing restores tissue integrity through fibrosis and scarring at a cost of regenerating the normal tissue architecture. Fetal cutaneous wounds made at certain developmental stages show complete regeneration. However, when fetuses are wounded later in gestation, dermal structures do not regenerate and a histologically demonstrable scar is formed (127, 128).

Kishi et al. (129) showed nerve regeneration in early gestational fetal wound healing was similar to unwounded skin. This re-innervation during healing appears to be by both collateral sprouting from intact nerves in the base of the wound and by regeneration of divided axons at the wound peripheries (130). However this regenerative capacity was lost as the wounding occurred at a later gestational age (131). This highlights the importance of cutaneous innervation in wound healing, specifically regeneration. In certain amphibious species such as newts, regeneration of amputated limbs is nerve dependant with regenerative ability lost upon denervation (131). Fetal cutaneous regeneration following wounding is also reported to be disrupted by denervation (132). Stelnicki et al. (132) showed that in fetal lambs whose limbs were denervated, incisional wounds appeared to scar, and open wounds failed to heal. The related dependence of scarless fetal repair and peripheral nerve function has also been shown in fetal mice where transection of intercostal nerves leads to loss of skin wound regeneration (129).

Previous studies (as explored in preceding sections) have focused on the role of neuromediators and their receptors in relation to adult cutaneous healing after injury. Similarly, during fetal injury this interaction of neuromediators may modulate the fetal scarless repair mechanisms in response to injury. SP and CGRP are undetectable during wound healing at early gestational age (130, 133). Zhang & Ren (134) showed SP expression altered from undetectable in early fetal skin to increasing expression with gestational term. Xie et al. (135) showed that fetal rabbit wounds healed more rapidly in comparison to adults without the formation of a scar. They showed the expression of SP and CGRP in the wound was decreased in the early stages of wound healing compared to fetal controls and remained at all times at levels equivalent to or below controls. These low levels of neuropeptide expression may contribute to scarless wound healing.

CONCLUSION AND FUTURE PERSPECTIVES

The skin is a densely innervated organ and the above clearly suggests that the cutaneous nervous system and its associated neuromediators are not solely responsible for sensory neurotransmissions to the central nervous system but play a major role in skin homeostasis and during all phases of wound healing. Delayed wound healing in clinical and experimental studies of denervated tissue have further supported this concept. This is highlighted with the imbalance of neuromediators found in denervated disease states such as diabetes, where delayed wound healing is common.

From this review it becomes apparent that pharmacological and mechanical methods of cutaneous re-innervation are an important avenue that needs to be explored further. One possibility is the use of electrical stimulation (ES) which has been shown to have beneficial effects in wound healing (136, 137). ES has been shown to enhance nerve regeneration (138, 139), however the mechanism by which it affects nerve growth and function is less understood. The functions of sensory nerves involved in wound healing have been shown to be improved by ES (140). A randomised controlled trial in humans showed ES improved factors known to reflect C-fibre function (141). The improvement in C-fibre function was associated with nearly doubling of the rate of healing in the actively stimulated group compared to the sham group. ES has been shown to improve regeneration of peripheral sensory axons both *in vitro* and *in vivo* (142–145). Another possible mechanism is that the use of ES may enhance the activity of neuromediators involved in wound healing (139, 140, 146). However the limited evidence available that shows the direct effect of electrical stimulation on skin innervation needs further clarification and verification.

Finally, the differing roles of cutaneous innervation and neuromediators between post natal and fetal wound healing may provide the opportunities to develop therapeutic technologies that could manipulate the central nervous system-skin peripheral nervous system network to not only aid in the management of delayed wound healing but also to allow cutaneous regeneration which would provide scarless healing.

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