Concise Review: Tissue-Engineered Skin and Nerve Regeneration in Burn Treatment

Mathieu Blais, Rémi Parenteau-Bareil, Sébastien Cadau, François Berthod

Key Words. Skin grafts • Tissue regeneration • Transplantation • Neuron • Epidermis • Cell biology

ABSTRACT

Burns not only destroy the barrier function of the skin but also alter the perceptions of pain, temperature, and touch. Different strategies have been developed over the years to cover deep and extensive burns with the ultimate goal of regenerating the barrier function of the epidermis while recovering an acceptable aesthetic aspect. However, patients often complain about a loss of skin sensation and even cutaneous chronic pain. Cutaneous nerve regeneration can occur from the nerve endings of the wound bed, but it is often compromised by scar formation or anarchic wound healing. Restoration of pain, temperature, and touch perceptions should now be a major challenge to solve in order to improve patients’ quality of life. In addition, the cutaneous nerve network has been recently highlighted to play an important role in epidermal homeostasis and may be essential at least in the early phase of wound healing through the induction of neurogenic inflammation. Although the nerve regeneration process was studied largely in the context of nerve transections, very few studies have been aimed at developing strategies to improve it in the context of cutaneous wound healing. In this concise review, we provide a description of the characteristics of and current treatments for extensive burns, including tissue-engineered skin approaches to improve cutaneous nerve regeneration, and describe prospective uses for autologous skin-derived adult stem cells to enhance recovery of the skin’s sense of touch.

INTRODUCTION

Although skin is the largest organ of the body, it is rarely seen as a vital one like the brain or the heart. However, a destruction of only 15% of the skin’s total body surface area is sufficient to be life-threatening. Indeed, the rupture of the skin’s barrier function induces massive water loss that will rapidly cause a deadly hypovolemic shock. In addition, burn is the most traumatic injury that the human body can bear and induces a complete disruption of the body’s homeostasis, including immunodepression, massive hypermetabolism, and vascular hyperpermeability, which enhances edema formation [1]. Major improvements in resuscitation techniques in the last 50 years enable the survival of patients with more than 90% of total body surface area burned. Then arises the challenge of covering these deep and extensive burns as fast as possible to protect the patients from septicemia and achieve the best functional and aesthetic healing possible.

There are approximately 15,000–20,000 hospitalizations per year for acute burn injuries in the U.S. recorded by the American Burn Association [2]. Most of them were caused by fire/flames, scald, electricity, and chemical products. The duration and intensity of exposure to these different factors, as well as location and depth of the trauma, determine the gravity of burn injuries. A first-degree burn is a superficial one that affects only the epidermis. Superficial second-degree burns also in part affect the dermis while preserving some epidermal rete ridges, enabling spontaneous scarless healing. Deep second-degree burns induce the complete destruction of epidermis but preserve hair follicles in the deep dermis from which epidermal healing can be initiated, and they promote hypertrophic scar formation.

Third-degree burns destroy both epidermis and dermis up to the hypodermis and necessitate a skin graft coverage. In a fourth-degree burn, underlying organs such as muscles, tendons, and bones are altered [2].

The gold standard to cover deep burns is to harvest split-thickness skin from unburned areas and graft it onto wounds. This will inflict a superficial second-degree burn at the donor site, which can heal in 2 weeks without scarring, while promoting good-quality coverage of the autograft skin on burns. However, the use of this method is limited if patients have few unburned areas left [1]. It necessitates a wait for the healing of donor sites and grafted wounds to harvest additional split-thickness...
skin. However, time is life for burn patients because uncovered wounds are at high risk of infection, highlighting the need for alternative burn coverage strategies.

**CHALLENGE OF TACTILE AND SENSORY RECOVERY**

The skin is a highly sensitive organ. It is densely innervated with different types of nerve endings, which discriminate between pain, temperature, and sense of touch. When a deep burn occurs, cutaneous nerves and their sensory corpuscles are destroyed while the sensory neuron cell bodies persist in the dorsal root ganglia along the spinal cord.

The cutaneous sensory nerves are broadly classified according to diameter and speed of impulse as Aβ, Aδ, and C nerve fibers, from the biggest and fastest to the smallest and slowest, respectively. They are associated with Schwann cells, which secrete a basal lamina around them and also produce myelin sheaths on the larger Aβ and Aδ fibers. Mechanical stimuli are detected via mechanoreceptors associated with sensory corpuscles through Aβ fibers (and approximately 20% of Aδ fibers), temperature via the thermoreceptors through Aδ and C fibers, and pain via the nociceptors through Aδ and C fibers (an extensive review on skin innervation is given in [3]).

After a deep burn injury, cutaneous nerve regeneration will occur with the migration of new nerve fibers from the wound bed or from the collateral sprouting of nerve fibers from the adjacent uninjured area. This nerve regeneration process is imperfect. It was reported that 71% of extensively burned victims suffer from abnormal sensations and 36% from chronic pain [4]. Victims often suffer permanent sensory deficits [5–7]. In addition, deficits are more pronounced if there was a skin graft [8], perhaps because these grafts were performed on the deepest burned areas. Among the possible causes of this abnormal sensitivity are the lack of nerve fibers [9, 10], a reinnervation mostly made of Aδ and C fibers with a deficit in Aβ fibers [11], and the lack of sensory units such as the sensory corpuscles and hair follicles [12]. Among other possible causes, there may be changes in the processing of information by the central nervous system [13, 14]. Hypersensitivity (allodynia), even in unburned parts of the body, may suggest that changes in the processing of sensory information by the central nervous system may contribute to some extent to the abnormal sensitivity [15].

**BASIC PRINCIPLES IN THE DEVELOPMENT OF A TISSUE-ENGINEERED SKIN**

The most promising technique to fulfill the clinical need to cover deep and extensive burns is to reconstruct a complete skin by tissue engineering using the patient’s own cells. The primary therapeutic concern is a rapid recovery of the skin barrier function to diminish water loss and prevent infection when the access to autografts is limited. This aim can be achieved by reconstruction of the epidermis. Most protocols to obtain autologous epidermis suitable for grafting from cultured keratinocytes are derived from the pioneering work of Rheinwald and Green in the mid-1970s [16]. To rebuild epidermal equivalents, keratinocytes purified from a small skin biopsy are grown on lethally irradiated murine 3T3 fibroblasts until they form a cell sheet. These 3T3 cells have an inhibitory influence on the growth of contaminating fibroblasts from the donor tissue. The addition to the medium of cholera toxin, which increases cellular cAMP, and of epidermal growth factor, which promotes proliferation, allows for a large-scale amplification of keratinocytes in vitro [17, 18].

Epithelial sheets can be directly grafted on donor sites to accelerate and improve their regeneration in order to harvest split-thickness autografts faster and several times from the same donor site. Alternatively, epithelial sheets can also be grafted alone on debrided burns, but with less optimal healing quality, or in combination with a widely meshed split-thickness autograft for a better aesthetic result [19–22].

However, the reconstruction of a complete tissue-engineered skin featuring both the epidermis and the dermis is the ultimate goal to improve healing quality and avoid scar formation. The stability of the attachment of the epidermis to a well-vascularized underlying dermis is a critical issue [23–26]. The dermis would also provide beneficial mechanical and paracrine support during the in vitro reconstruction and the transplantation.

But whereas the keratinocytes naturally organize in basal and suprabasal cell layers when cultured on a plastic dish, fibroblasts do not, in conventional culture conditions. For that reason, most methods developed to reconstruct a dermis rely on the culture of fibroblasts in a scaffold to build a three-dimensional tissue.

The biomaterial used in the dermal portion can be natural, synthetic, or both and made of autologous or allogenic fibroblasts [27]. Type I and III collagens being the major proteins of the skin extracellular matrix (ECM), they were widely used to reconstruct dermal compartments. The first full-thickness skin was reconstructed using a fibroblast-populated collagen gel seeded with keratinocytes [28]. Several biomaterials made of collagens or biodegradable polymers were marketed for the reconstruction of the skin, notably collagen sponges [29]. An alternative method, a dermal construct made of an ECM secreted and assembled by the fibroblasts themselves was developed after addition of ascorbate to the culture medium [30]. Since fibroblasts synthesize collagen naturally, this feature can be exploited to produce fibroblast sheets that can be stacked together to achieve thicker dermal compartment prior to keratinocytes seeding [31]. This method enables the production of a tissue-engineered skin exclusively made of the patient’s cells.

**OUR APPROACH TO ENHANCE CUTANEOUS NERVE REGENERATION**

After the destruction of nerve endings following deep burns, axonal migration can occur from the wound bed into the healed skin. However, this process takes place in the absence of the critical guidance cues derived from cutaneous neuroanatomical structures such as hair follicles and sensory corpuscles destroyed in deep burns [32]. In addition, anarchic remodeling of the dermal ECM during wound healing may impair nerve migration. If most current strategies to reconstruct skin result in the development of tissues without appendage, a well-structured ECM may facilitate tissue innervation.

The graft of a sponge biomaterial made of collagen and chondroitin sulfate in a cutaneous wound has been shown to enhance nerve regeneration from the edges. This effect was mostly attributed to a decrease of wound contraction and fibrosis promoted by the biomaterial [33]. A tissue-engineered skin made of a collagen hydrogel combined with fibroblasts and keratinocytes, keratinocytes and melanocytes, or sweat glands was grafted on rats for 3 and 8 weeks. Some nerve ingrowth was observed in all
three constructs after only 8 weeks, limited to the dermis [34]. A human skin explant has also been cultured in vitro for 10 days and combined with mouse sensory neurons to analyze interactions between skin and nerves [35].

In our group, we investigated whether a tissue-engineered skin could be innervated after graft in mice. We used a reconstructed skin made of human keratinocytes seeded on a collagen sponge populated with fibroblasts promoting the deposition and remodeling of a highly physiological ECM [36, 37]. We first showed that this connective tissue facilitates axonal migration in vitro by seeding dorsal root ganglia-derived mouse sensory neurons at the bottom of the tissue-engineered skin (Fig. 1B) [38]. When the reconstructed skin was transplanted on immunodeficient mice, nerve fibers from the wound bed were observed to migrate into the transplant after 2 months [39]. Schwann cells were also detected migrating in the graft and forming a Büngner band-like structure to guide axonal migration [39]. These results showed that a well-structured tissue-engineered skin can enhance nerve regeneration.

Schwann cells and the basement membrane components they produce, such as laminin, appear to play a critical role in peripheral nerve regeneration [40]. We investigated whether nerve regeneration could be improved by adding laminin to the collagen sponge used to reconstruct skin [41]. Four months after transplantation, there were seven times as many neurofilament M-positive nerve fibers migrating in the graft in the condition enriched with 10 μg of laminin per sponge. There was also a significant improvement in the current perception threshold of the transplant for the Aβ and Aδ nerve fibers in all grafts enriched with laminin with respect to the control. In addition, whereas a larger number of nerve fibers was observed in the laminin group compared with normal mouse skin, it did not induce skin hyperesthesia [41]. These results showed that it is possible to increase nerve migration in a tissue-engineered organ using laminin.

Since laminin can be secreted by Schwann cells and these cells are major contributors to the nerve regeneration process, we hypothesized that the addition of Schwann cells in our tissue-engineered skin could also promote nerve regeneration. We extracted Schwann cells from mouse peripheral nerves and incorporated them with the fibroblasts in our reconstructed skin.
model innervated in vitro with mouse sensory neurons. Schwann cells spontaneously localized along nerve fibers (Fig. 1C, 1D) and achieved the formation of myelin sheaths around axons in vitro as assessed by transmission electron microscopy [42]. In addition, they promoted a twofold increase in the number of sensory fibers migrating in the reconstructed skin as compared with the control without Schwann cells. Once our tissue-engineered skin was transplanted on nude mice, Schwann cells induced a 1.8-fold increase in the number of nerve fibers migrating in the graft 60 days after transplantation, and promoted a sensory recovery of the transplant with a current perception threshold similar to that of normal skin for the large and myelinated Aβ-sensory fibers (responsible for the sense of touch), in contrast with transplants without Schwann cells [42]. Overall, the addition of laminin or Schwann cells in reconstructed skin promoted a significant improvement in nerve regeneration for the Aβ and Aδ nerve fibers. This improvement is interesting since there is a deficit in the regeneration of these large nerve fibers in deep and extensive burns after skin grafts [11]. However, the Aδ fibers will not be able to detect sense of touch perception as long as they are not connected to a tactile sensor such as a sensory corpuscle (Meissner, Pacini, Ruffini, and Merkel touch dome) or a hair follicle. Although how to reconstruct a corpuscle remains largely unknown, attempts to incorporate hair follicles in tissue-engineered skin have already been investigated [43]. Using a tissue-engineered skin with mouse hair bud-like structures, we analyzed the impact of these appendages on axonal migration. We showed that hair bud-like structures established in vitro specific guiding cues to promote a preferential axonal migration around them, but failed to make mature hairs. When this reconstructed skin was transplanted on nude mice, hair buds successfully developed into hairs, and their presence accelerated nerve regeneration (occurring after 1 month instead of 2 months) and oriented it to preferentially innervate hair shafts (Fig. 1E) [44]. This result showed that it is possible to orient nerve regeneration in a tissue-engineered organ using specific cells that can establish neuronal guiding cues.

SKPs derive from the neural crest and are located mainly in the interfollicular dermal papillae (in glabrous skin) and the hair follicle bulge region where they contribute to the homeostasis of the skin, the hair follicles, and the sebaceous glands in the adult [48–50]. We and others successfully isolated these precursors from adult human skin biopsies and demonstrated their potential to differentiate into mature neurons [51]. These precursors were also shown to have the potential to differentiate into Schwann cells [52].

Multipotent mesenchymal stem cells are present in the dermis in proximity of hair follicles in perivascular sites that may act as a niche in human scalp skin [53]. However, a growing body of evidence suggests that pericytes, the mural cells located around small capillaries in connective tissues, may bear multipotent stem cell properties [54]. Thus, SKPs and MSCs should be viewed as two different sources of stem cells, SKP location being restricted to epidermis and hair follicle, with a main role to play in skin homeostasis. In contrast, MSCs are distributed in the whole dermis around capillaries and may be devoted to the regeneration of more distant and various tissues, taking advantage of their unique position close to blood circulation [55]. Dermal MSCs should be identical to muscle or adipose-derived MSCs, with a similar location. Finally, isolation of skin-derived stem cells from full skin biopsies may correspond to a mixed population of SKPs and MSCs.

Adipose-derived stem cells are multipotent cells characterized by the expression of mesenchymal stem cells markers. They are present in the hypodermis, and they can be enriched in cell culture from the stromal vascular fraction of homogenized fat tissue [56]. The use of stem cells from adipose tissue is advantageous because they can be obtained by liposuction, a common and minimally invasive procedure. In addition, this cell population is abundant and has already been grafted in humans [56]. Their potential of differentiation into Schwann cell has also been demonstrated [57].

Developing a tissue-engineered skin with hair follicles would be a promising approach to enhance sense of touch recovery after transplantation. However, generating hair follicles from the differentiation of stem cells in vitro is a challenge for functional skin reconstruction. The addition of hair follicles would not only improve nerve regeneration but would also provide a niche for stem cells involved in hair renewal and wound healing [58, 59]. De novo formation of hair follicle in vivo was shown to be possible using epithelial and mesenchymal cells from embryos as well as newborn mice or stem cells from the adult bulge area [60, 61].

Recently, the regeneration of both pelage hair follicle and vibrissae from suspensions of dorsal embryonic skin plus epidermal bulge cells and dermal papilla cells cultured under the subcapsule was shown in an elegant study [62–64]. Long-term cultured dermal papilla cells from vibrissae are also able to induce de novo hair follicle development in athymic mice without cell suspension [65]. Tissue-engineered skin with hair follicles could be successfully obtained after its engraftment using neonatal multipotent skin precursor cells [66]. The most advanced in vitro model for hair follicle regeneration is based on dermal papilla cells. These cells are cultured in aggregate, and then keratinocytes from outer root sheath and hair follicle melanocyte are added to form microfollicles in vitro that result in hair shaft formation [67]. Taken together, these data show that stem cells from the bulge area or from dermal papilla as well as embryonic back skin suspension cells have the ability to induce hair in vitro. It should, therefore, be possible to obtain hair shafts in reconstructed skin in vitro, prior to the graft.

©AlphaMed Press 2013

Cutaneous Nerve Regeneration
After the clinical success of autologous epithelial sheets for the coverage of deep and extensive burns, the next step will be the transplantation of patient-derived tissue-engineered skin that promotes a better healing quality. Now that the essential cutaneous barrier function can be recovered with acceptable aesthetic outcome, the question of how improving the perceptions of pain, temperature, and sense of touch of these large skin areas is arising. Different strategies can be followed, from the easiest to the most complex (Fig. 2). At the beginning, a well-structured connective tissue ECM is the basis to facilitate nerve migration while avoiding scar formation from the wound bed. Then, the simple combination into the transplant of highly stable and large molecules such as laminin, which are known to enhance axonal migration, could be an easy way to ensure a general, but not specifically guided, nerve regeneration. A more complex approach relying on the isolation and differentiation of patient-derived stem cells into Schwann cells prior to being included in the tissue-engineered skin in order to promote nerve regeneration. Laminin can be embedded into a biomaterial subsequently used to prepare the tissue-engineered skin. Both approaches promote pain and temperature perceptions but not recovery of the sense of touch. Any of these possibilities will promote a better nerve regeneration in grafted skin compared with the same tissue made only of fibroblasts and keratinocytes, which will fail to achieve pain and temperature perceptions identical to those of normal skin, and will not promote sense of touch recovery. In contrast, late coverage of wounds without dermal regeneration will induce scar formation and loss of touch, pain, and temperature perceptions, in addition to increased risks of chronic pain and pruritus. Abbreviations: ORS, outer root sheath; TE, tissue-engineered.

CONCLUSION

Figure 2. Schematic representation of different strategies to enhance nerve regeneration in a tissue-engineered skin. All the cellular components should preferentially be isolated from a single uninjured autologous skin biopsy. Dermal papilla cells cultured with ORS keratinocytes from hair follicles and melanocytes (according to the differentiation method of Lindner et al. [67]) can be added to promote hair bud formation and growth, and functional mecanoeceptor regeneration. Hairs may also be obtained from skin- or adipose-derived stem cells. This approach would enhance touch, pain, and temperature perceptions. Skin-derived precursor cells or adipose-derived mesenchymal stem cells can be differentiated into Schwann cells prior to being included in the tissue-engineered skin in order to promote nerve regeneration. Laminin can be embedded into a biomaterial subsequently used to prepare the tissue-engineered skin. Both approaches promote pain and temperature perceptions but not recovery of the sense of touch. Any of these possibilities will promote a better nerve regeneration in grafted skin compared with the same tissue made only of fibroblasts and keratinocytes, which will fail to achieve pain and temperature perceptions identical to those of normal skin, and will not promote sense of touch recovery. In contrast, late coverage of wounds without dermal regeneration will induce scar formation and loss of touch, pain, and temperature perceptions, in addition to increased risks of chronic pain and pruritus. Abbreviations: ORS, outer root sheath; TE, tissue-engineered.
tissue-engineered skin and should be very promising in terms of sense of touch recovery in addition to an improvement of the aesthetic aspect of the skin.

ACKNOWLEDGMENTS

This work was supported by the Canadian Institutes of Health Research (Grant MOP-106429). M.B. is a recipient of a doctoral scholarship from the Fonds de Recherche du Québec en Santé.

REFERENCES

1. Berthod F, Rouabhia M. Exhaustive review of clinical alternatives for damaged skin replacement. In: Rouabhia M, ed. Skin Substitute Production by Tissue Engineering: Clinical and Fundamental Applications. Austin, TX: Landes Bioscience 1997:23–45.

2. Mendez-Eastman S. Burn injuries. Plast Surg Nurs 2005;25:133–139.

3. Roosterman D, Goerge T, Schneider SW et al. Neuronal control of skin function: The skin as a neuroimmunoendocrine organ. Physiol al. Neuronal control of skin function: The skin as a neuroimmunoendocrine organ. Physiol 1987;211:1052–1053.

4. Malenfant A, Forget R, Papillon J et al. Prevalence and characteristics of chronic sensory problems in burn patients. Pain 1996;67:493–500.

5. Hermanson A, Jonsson CE, Lindblom U. Sensitivity after burn injury. Clin Physiol 1986; 5:507–521.

6. Ward RS, Saffle JR, Schnebly WA et al. Sensory loss over grafted areas in patients with burns. J Burn Care Rehabil 1989;:10:536–538.

7. Ward RS, Tuckett RP. Quantitative thresholds of cutaneous sensation of patients with burns. J Burn Care Rehab 1991;12:569–575.

8. Malenfant A, Forget R, Amsel R et al. Tactile, thermal and pain sensitivity in burned patients with and without chronic pain and paresthesia problems. Pain 1998;77:241–251.

9. Altun V, Hakvoort TE, van Zuijlen PP et al. Nerve outgrowth and neuropathic expression during the remodeling of human burn wound scars: A 7-month follow-up study of 22 patients. Burns 2001;27:717–722.

10. Stella M, Calcagni M, Teich-Alasia S et al. Sensory endings in skin grafts and scars after extensive burns. Burns 1994;20:491–495.

11. Ward RS, Tuckett RP, English KB et al. Substance P axons and sensory threshold increase in burn-graft human skin. J Surg Res 2004;118:154–160.

12. Nedelee B, Hou Q, Sohbi I et al. Sensory perception and neuroanatomical structures in normal and grafted skin of burn survivors. Burns 2005;31:817–830.

13. Coderre TJ, Melzack R. Increased pain sensitivity following heat injury involves a central mechanism. Behav Brain Res 1985;15:259–262.

14. Coderre TJ, Melzack R. Cutaneous hyperalgesia: Contributions of the peripheral and central nervous systems to the increase in pain sensitivity after injury. Brain Res 1987;404:95–106.

15. Sang CN, Gracely RH, Max MB et al. Capsaicin-evoked mechanical allodynia and hyperalgesia cross nerve territories: Evidence for a central mechanism. Anesthesiology 1996;85:491–496.

16. Green H, Kehinde O, Thomas J. Growth of cultured human epidermal cells into multiple epithelia suitable for grafting. Proc Natl Acad Sci USA 1979;76:5665–5668.

17. Rheinwald JG, Green H. Epidermal growth factor and the multiplication of cultured human epidermal keratinocytes. Nature 1977;265:421–422.

18. Green H. Cyclic AMP in relation to proliferation of the epidermal cell: A new view. Cell 1978;15:801–811.

19. Gallico GG 3rd, O’Connor NE, Compton CC et al. Permanent coverage of large burn wounds with autologous cultured human epithelium. N Engl J Med 1984;311:448–451.

20. De Luca M, Albanese E, Bondanza S et al. Multicentre experience in the treatment of burns with autologous and allogeneic cultured epithelium, fresh or preserved in a frozen state. Burns 1989;15:303–309.

21. Donati L, Magliacani G, Borniolli M et al. Clinical experiences with keratinocyte grafts. Burns 1992;18(suppl 1):S19–S26.

22. Gallico GG 3rd, O’Connor NE. Cultured epithelium as a skin substitute. Clin Plast Surg 1985;12:149–157.

23. Cuono C, Langdon R, McGuire J. Use of cultured epidermal autografts and dermal allografts as skin replacement after burn injury. Lancet 1986;1:1123–1124.

24. Michel M, L’Heureux N, Pouliot R et al. Characterization of a new tissue-engineered human skin equivalent with hair. In Vitro Cell Dev Biol Anim 1999;35:318–326.

25. Boyce ST, Goretsky MJ, Greenhalgh DG et al. Comparative assessment of cultured skin substitutes and native skin autograft for treatment of full-thickness burns. Ann Surg 1995; 222:743–752.

26. Clugston PA, Snelling CF, Macdonald IB et al. Cultured epithelial autografts: Three years of clinical experience with eighteen patients. J Burn Care Rehabil 1991;12:533–539.

27. Place ES, Evans ND, Stevens MM. Complexity in biomaterials for tissue engineering. Nat Mater 2009;8:457–470.

28. Bell E, Ihrlich HP, Buttke DJ et al. Living wound tissue can utilize a polymeric template to synthesize a functional extension of skin. Science 1982;215:174–176.

29. Pouliot R, Larouche D, Auger FA et al. Reconstructed human skin produced in vitro and grafted on athymic mice. Transplantation 2002;73:1751–1757.

30. Larouche D, Paquet C, Fradette J et al. Regeneration of skin and cornea by tissue engineering. Meth Mol Biol 2009;482:233–256.

31. Peters EM, Botchkarev VA, Muller-Rover S et al. Developmental timing of hair follicle and dorsal skin innervation in mice. J Comp Neurol 2002;448:28–52.

32. Soller EC, Tzerinis DS, Miu K et al. Common features of optimal collagen scaffolds that disrupt wound contraction and enhance regeneration both in peripheral nerves and in skin. Biomaterials 2012;33:4783–4791.

33. Biedermann T, Botthcher-Haberzeth S, Klar A et al. Rebuild, restore, reinnervate: Do human tissue engineered dermo-epidermal skin analogs attract host nerve fibers for innervation? Ped Surg Int 2013;29:71–78.

34. Lebonvallet N, Boulais N, Le Gall C et al. Effects of the re-innervation of organotypic skin explants on the epidermis. Exp Dermatol 2012;21:156–158.

35. Berthod F, Germain L, Guignard R et al. Differential expression of collagens XII and XIV in human skin and in reconstructed skin. J Invest Dermatol 1997;108:737–742.

36. Berthod F, Germain L, Li H et al. Collagen fibril network and elastic system remodeling in a reconstructed skin transplanted on nude mice. Matrix Biol 2001;20:463–473.

37. Jingras M, Bergeron J, Dery J et al. In vitro development of a tissue-engineered model of peripheral nerve regeneration to study neurite growth. FASEB J 2003;17:2124–2126.

38. Jingras M, Paradis I, Berthod F. Nerve regeneration in a collagen-chitosan tissue-engineered skin transplanted on nude mice. Biomaterials 2003;24:1653–1661.

39. Høkå A. Mechanisms of disease: What factors limit the success of peripheral nerve regeneration in humans? Nat Clin Pract Neuro 2006;2:448–454.

40. Caisse R, Jingras M, Champigny MF et al. In vivo enhancement of sensory perception recovery in a tissue-engineered skin enriched with laminin. Biomaterials 2006;27:2988–2993.

41. Blais M, Grenier M, Berthod F. Improvement of nerve regeneration in tissue-engineered skin enriched with Schwann cells. J Invest Dermatol 2009;129:2895–2900.

42. Larouche D, Cuffley K, Paquet C et al. Tissue-engineered skin preserving the potential of epithelial cells to differentiate into hair after grafting. Tissue Eng Part A 2011;17:819–830.

43. Gagnon V, Larouche D, Parenteau-Bareil R et al. Hair follicles guide nerve migration in vitro and in vivo in tissue-engineered skin. J Invest Dermatol 2011;131:1375–1378.

AUTHOR CONTRIBUTIONS

M.B.: conception and design, manuscript writing, collection and assembly of data; R.P.-B.: manuscript writing, collection and assembly of data; S.C.: manuscript writing; F.B.: conception and design, manuscript writing, financial support, final approval of manuscript.

DISCLOSURE OF POTENTIAL CONFLICTS OF INTEREST

The authors declare no potential conflict of interest.

550 Cutaneous Nerve Regeneration

©AlphaMed Press 2013
Sipp D. Challenges in the clinical application of induced pluripotent stem cells. Stem Cell Res Ther 2010;1:9.

Blanpain C, Fuchs E. Epidermal homeostasis: A balancing act of stem cells in the skin. Nat Rev Mol Cell Biol 2009;10:207–217.

Nishimura EK. Melanocyte stem cells: A melanocyte reservoir in hair follicles for hair and skin pigmentation. Pigment Cell Melanoma Res 2011;24:401–410.

Fernandes KJ, McKenzie IA, Mill P et al. A dermal niche for multipotent adult skin-derived precursor cells. Nat Cell Biol 2004;6:1082–1093.

Biernaskie J, Paris M, Morozova O et al. SKPs derive from hair follicle precursors and exhibit properties of adult dermal stem cells. Cell Stem Cell 2009;5:610–623.

Hunt DP, Morris PN, Sterling J et al. A highly enriched niche of precursor cells with neuronal and glial potential within the hair follicle dermal papilla of adult skin. Stem Cells 2008;26:163–172.

Gingras M, Champigny MF, Berthod F. Differentiation of human adult skin-derived neuronal precursors into mature neurons. J Cell Physiol 2007;210:498–506.

McKenzie IA, Biernaskie J, Toma JG et al. Skin-derived precursors generate myelinating Schwann cells for the injured and dysmyelinated nervous system. J Neurosci 2006;26:6651–6660.

Yamanishi H, Fujiwara S, Soma T. Perivascular localization of dermal stem cells in human scalp. Exp Dermatol 2012;21:78–80.

Bouacida A, Rosset P, Trichet V et al. Pericyte-like progenitors show high immaturity and engraftment potential as compared with mesenchymal stem cells. PLoS One 2012;7:e48648.

Ema H, Suda T. Two anatomically distinct niches regulate stem cell activity. Blood 2012;120:2174–2181.

Tobita M, Orbay H, Mizuno H. Adipose-derived stem cells: Current findings and future perspectives. Discov Med 2011;11:160–170.

Kaewkhaw R, Scott AM, Haycock JW. Anatomical site influences the differentiation of adipose-derived stem cells for Schwann-cell phenotype and function. Glia 2011;59:734–749.

Itô M, Yang Z, Andl T et al. Wnt-dependent de novo hair follicle regeneration in adult mouse skin after wounding. Nature 2007;447:316–320.

Oshima H, Rochat A, Kedzia C et al. Morphogenesis and renewal of hair follicles from adult multipotent stem cells. Cell 2001;104:233–245.

Zheng Y, Du X, Wang W et al. Organogenesis from dissociated cells: Generating of mature cycling hair follicles from skin-derived cells. J Invest Dermatol 2005;124:867–876.