

# Inflammation and cutaneous nervous system involvement in hypertrophic scarring

Shao-hua Li<sup>1</sup>, Heng-lian Yang<sup>2</sup>, Hu Xiao<sup>1,\*</sup>, Yi-bing Wang<sup>1</sup>, De-chang Wang<sup>1</sup>, Ran Huo<sup>1</sup>

1 Shandong Provincial Hospital Affiliated to Shandong University, Jinan, Shandong Province, China

2 Jinan Fire Protection Hospital, Jinan, Shandong Province, China

**\*Correspondence to:**

Hu Xiao, Ph.D., 847247374@qq.com.

**orcid:**

0000-0002-4058-5782 (Hu Xiao)

**doi:** 10.4103/1673-5374.167769

**http://www.nrronline.org/**

**Accepted:** 2015-06-15

## Abstract

This study aimed to use a mouse model of hypertrophic scarring by mechanical loading on the dorsum of mice to determine whether the nervous system of the skin and inflammation participates in hypertrophic scarring. Results of hematoxylin-eosin and immunohistochemical staining demonstrated that inflammation contributed to the formation of a hypertrophic scar and increased the nerve density in scar tissue. Western blot assay verified that interleukin-13 expression was increased in scar tissue. These findings suggest that inflammation and the cutaneous nervous system play a role in hypertrophic scar formation.

**Key Words:** nerve regeneration; peripheral nerve regeneration; hypertrophic scar; interleukin-13; wound healing; nerve growth factor; neural regeneration

**Funding:** This study was supported by a grant from the Development of Medical Science and Technology Project of Shandong Province in China, No. 2014WS0354; the National Natural Science Foundation of China, No. 81272099; the Shandong Medical and Health Science and Technology Development Program Fund for Youth in China, No. 2009QZ023; and the Natural Science Foundation of Shandong Province in China, No. BS2009YY043.

Li SH, Yang HL, Xiao H, Wang YB, Wang DC, Huo R (2015) Inflammation and cutaneous nervous system involvement in hypertrophic scarring. *Neural Regen Res* 10(10):1678-1682.

## Introduction

Hypertrophic scars and keloids form as a result of aberrations of physiologic wound healing and may arise following any insult to the deep dermis. These fibroproliferative diseases of the skin present a major therapeutic dilemma and challenge to the plastic surgeon because they are disfiguring and frequently recur (Perry et al., 2010). During the past several decades, many studies have been performed to determine the etiology of hypertrophic scarring (Bloemen et al., 2009; Henderson et al., 2011; Atiyeh and Janom, 2014). Several studies have shown that neural factor is an important molecule that promotes hypertrophic scarring (Citak et al., 2010; Feng et al., 2010; Hamed et al., 2011; Yagmur et al., 2011; Henderson et al., 2012). Earlier studies suggested a relation between blood levels of the inflammatory mediator interleukin-13 (IL-13) and the progression of wound healing, which resulted in further tissue destruction and finally, scar formation (Akaishi et al., 2008; Ngeow, 2010; Buckley et al., 2011; Park et al., 2011; Elsharawy et al., 2012). This study investigated the relationship between inflammation and skin nerve regeneration with hypertrophic scarring in a mouse model of hypertrophic scarring.

## Materials and Methods

### Establishment of scar models

Sixty clean, female C57BL/6 mice, aged 8 weeks old and weighing  $20 \pm 5$  g, were provided by the Laboratory Animal

Center of Shandong University in China (license No. SCXK (Lu) 2003-0004). They were administered water and food regularly and had no adverse factors. The protocols were consistent with the required animal ethics requirements. This research received permission of the Medical Ethics Committee of Shandong Provincial Hospital affiliated to Shandong University, China. Mice were equally and randomly assigned to either the control or experimental groups.

The mice were anesthetized by intraperitoneal injection of 3% pentobarbital (1 mL/kg). The model of hypertrophic scar was established as previously described (Aarabi et al., 2007). In brief, biomechanical loading devices were constructed from 22-mm expansion screws (Great Lakes Orthodontic Products, Tonawanda, NY, USA). A median incision approximately 2-cm long was made on the dorsum of the mouse, then sutured with 6-0 nylon sutures. At 4 days following injury, the sutures were removed from the scars, and a loading device was carefully fixed with 6-0 nylon sutures. Wounds in the experimental group were loaded every other day. Prior to applying tension, two points were identified on either side of the scars using a permanent marking pen. Tension on the wounds was created by carefully distracting the expansion screws by 2 mm at 4 days after injury and 4 mm every other day thereafter. During the periods between distractions, stress relaxation was observed due to the natural elongation of skin resulting in a continuous decrease in the force acting on the wounds. To compensate, tension was reapplied every other day for up to 2 weeks. The loading devices

of the control group were not activated. The backs of the animals were shaved prior to morphometric measurements. Scar tissue was harvested at 2 and 5 weeks following the initiation of strain (15 mice at each time point in each group).

### Hematoxylin-eosin staining

Five- $\mu\text{m}$ -thick sections were dewaxed using xylene and washed with alcohol and water, followed by hematoxylin staining for 5 minutes. Sections were washed with tap water, differentiated with hydrochloric acid for 30 seconds, soaked in tap water for 15 minutes, and stained with eosin for 2 minutes. Sections were then dehydrated, cleared and mounted. Microstructural changes in tissues were observed by light microscopy (Type BX51, Olympus, Tokyo, Japan).

### Immunohistochemical staining

The specimens were embedded in paraffin and cut into 4- $\mu\text{m}$ -thick sections for immunohistochemical staining. Tissue sections were deparaffinized and microwaved for antigen retrieval, followed by incubation in 3%  $\text{H}_2\text{O}_2$  for 10 minutes. Nonspecific binding of antibodies was inhibited by incubation in 5% normal goat serum for 20 minutes in a humidified chamber. Sections were washed with PBS three times, and incubated with rabbit anti-S100 polyclonal antibody (1:100; Abcam, Cambridge, MA, USA) for 40 hours at 4°C. After three washes with PBS, tissue sections were incubated with biotinylated goat anti-rabbit IgG (1:1,000; Boster, Wuhan, Hubei Province, China) for 30 minutes at room temperature. After washing with PBS, slides were incubated in streptavidin-peroxidase complex for 30 minutes at 37°C, washed three times, visualized using diazaminobenzene, and counterstained with hematoxylin. As a negative control, sections were stained without the addition of a primary antibody. Expression of S100 was quantitatively analyzed using morphological analysis software (Qwind Pro V3.3.1, Leica, Germany). All measurements were assessed by physicians blind to the group being studied.

### Western blot assay

Total protein was extracted from scar tissue samples using T-PER Tissue Protein Extraction Reagent (Pierce, Rockford, IL, USA). Protein samples (20  $\mu\text{g}$ /lane) were mixed with 5- $\mu\text{L}$  loading buffer (50 mM Tris HCl, pH 6.8, 2% sodium dodecyl sulfate, 0.1% bromophenol blue, 10% glycerol) and 2-mercaptoethanol (96 mM) and heated for 5 minutes. Lysates were fractionated on sodium dodecyl sulfate polyacrylamide gel electrophoresis and transferred onto a nitrocellulose membrane (Bio-Rad). The nitrocellulose membrane was blocked for 2 hours at 4°C in blocking solution containing 3% bovine serum albumin in 1  $\times$  Tween 20 Tris buffered saline, supplemented with 20 mM Tris HCl, pH 7.4, 150 mM NaCl, and 0.1% Tween 20. After blocking, the membrane was incubated with rabbit anti-IL-13 polyclonal antibody (1:1,000; Abcam), rabbit anti-mouse beta-actin monoclonal antibody (1:5,000; Sigma-Aldrich, St. Louis, MO, USA) and incubated for 2 hours at 37°C with gentle shaking on a platform shaker. After incubation with goat anti-rabbit IgG (1:2,000; Santa Cruz Biotechnology, Santa Cruz, CA, USA). Expression was

detected by enhanced chemiluminescence kit (Amersham Biosciences, Little Chalfont, Buckinghamshire, UK). Film autoradiograms were exposed for 10–30 minutes. The absorbance values of the target protein were measured using the image analysis program Image J (Toronto Western Research Institute University Health Network, Toronto, ON, Canada). Beta-actin was used as an internal loading control. The optical density ratio of target protein to internal reference was represented as the relative amount of target protein.

### Statistical analysis

Data were analyzed by SPSS 10.0 statistical analysis software (SPSS, Chicago, IL, China), and experimental data are expressed as the mean  $\pm$  SEM. The differences between the two groups were analyzed by independent samples *t*-test.  $P < 0.05$  was considered statistically significant.

## Results

### Histological observation of scars in a mouse model of hypertrophic scarring

Similar to human hypertrophic scars, the scars in mice in the experimental group were increased in numbers and showed epidermal thickening with adnexal structures and hair follicles were absent in the dermis (**Figure 1A**). In all cases, the control wounds healed with almost no scarring (**Figure 1B**).

### Skin nerves in a mouse model of hypertrophic scarring

S-100-immunoreactive nerve fibers, brown and shaped as dots, short bars or bean sprouts, were visible in the scar tissue layer in the experimental group (**Figure 2A**). The control wounds demonstrated almost no S-100 Schwann cells (**Figure 2B**). The nerve fiber density in the experimental group was increased significantly compared with the control group ( $P < 0.05$ ; **Figure 2C**).

### Increased IL-13 expression in scars in a mouse model of hypertrophic scarring

IL-13 is an important pro-fibrotic cytokine for wound healing and scarring (Mandal et al., 2010; Wong et al., 2011). A western blot assay was used to measure IL-13 production in scars. At 2 weeks after injury, IL-13 levels in the experimental group increased significantly compared with the controls ( $P < 0.05$ ).

At 5 weeks, IL-13 levels in the experimental group remained significantly elevated above that of the control group. There were no significant differences in IL-13 levels between the two time points, at 2 and 5 weeks after injury in the experimental group (**Figure 3**).

## Discussion

Healed partial thickness wounds including burns and donor sites cause hypertrophic scar formation and patient discomfort. Although numerous factors are implicated in skin fibrosis, the exact pathophysiology of hypertrophic scarring remains unknown. Potential etiologies thought to underlie human hypertrophic scar and keloid formation include neural factors and inflammation (Scott et al., 2007; Pradhan

et al., 2009; Stein and Kuchler, 2013). A large number of previous studies showed that neural factors are important in regulating wound healing; however, the relationship between neural regulation and scar formation after wound healing is poorly understood (Pradhan et al., 2009; Khattak et al., 2010; Anderson et al., 2011; Isoardo et al., 2012; Nakagaki et al., 2013).

Increasing data suggest that the skin nervous system is involved in wound healing, and that its innervation is related to physiological functions and pathological changes of the skin (Moore et al., 2009; Buckley et al., 2011; Rashmi et al., 2012; Yasui et al., 2012). Recent studies demonstrated that peripheral nerves are closely related to the progress of scarless fetal wound healing (Antony et al., 2010; Yates et al., 2012; Akita et al., 2013; Hu et al., 2014; Liu et al., 2014; Morris et al., 2014). Mammoto et al. (2008) suggested that local application of nerve growth factor improved ligament healing by promoting both reinnervation and angiogenesis, and resulted in the formation of scars with enhanced mechanical properties. Wang et al. (2010) suggested that hypertrophic scars exhibited a greater number of nerve fibers with more serious pathologies compared with mature scars. However, the remodeling of regenerating nerve fibers during wound healing was observed, which changed nerve innervation density (Feng et al., 2010; Ishiguro et al., 2010; Anderson et al., 2011; Kim et al., 2011).

Most previous studies focused on the function of neural factors and wound healing speed (Stout, 2010; Sun et al., 2010; Tan et al., 2010; Yasui et al., 2012; Shen et al., 2013). However, there have been few reports regarding the relationship between neural factors and inflammation. Collagen production is down regulated by pro-inflammatory cytokines such as interferon- $\gamma$  and tumor necrosis factor- $\alpha$  (Anderson et al., 2011; Soller et al., 2012; Reichard et al., 2014). While the causes of fibrosis are unknown, inflammatory mediators such as IL-13 that stimulate collagen production have been implicated in the disease process. The stimulation of collagen production in fibroblasts is dependent on Smad and STAT6 signaling pathways induced by tumor growth factor- $\beta$  and IL-13, respectively (Malin et al., 2009; Zhang et al., 2010; Romano et al., 2014). However, these inflammatory mediators have not been investigated in tissues locally at the site of mechanical loaded trauma (Spits and Di Santo, 2011; Kumar, 2014).

This study used a mouse model of hypertrophic scars that are grossly and histologically identical to human hypertrophic scars, produced by applying exogenous mechanical forces onto healing mouse wounds. Our study showed that IL-13 levels were increased in scar tissue induced by mechanical strain. These findings are consistent with previous histological findings of increased IL-13 levels during wound healing process (Barron and Wynn, 2011; Nguyen et al., 2011; Ferrante and Leibovich, 2012; Gaspar et al., 2013; Hofmann et al., 2014). Although acute inflammation is necessary for normal wound repair, chronic inflammation can lead to pathological fibrosis (Semlali et al., 2010; Wang and Zhao, 2010; Oh et al., 2011; Novak and Koh, 2013; Weissner et al., 2013; Wise et al., 2013; Niu et al., 2014). Our study suggested that hypertrophic scars induced by mechanical strain

exhibited a greater number of nerve fibers, with more serious pathologies compared with control scars. Furthermore, mechanical forces modulate wound repair and hypertrophic scarring *via* neurogenic inflammatory pathways in tissues.

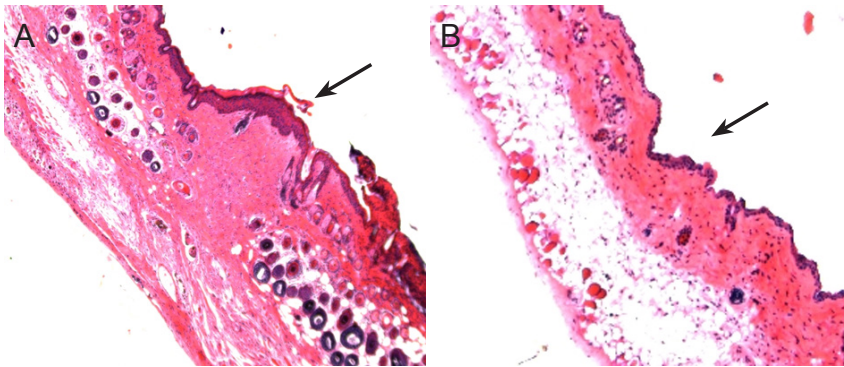
The association between neural factors and inflammation in hypertrophic scars is a novel finding, as there have been no previous reports of this relationship. Here, we demonstrate that focusing on the neural inflammatory pathways involved in skin fibrosis might be a useful strategy to identify new therapeutic approaches for the treatment of hypertrophic scars.

**Author contributions:** HX was a funding manager. YBW and DCW participated in laboratory operation, data recording and paper writing. RH, HLY and SHL participated in data analysis. All authors approved the final version of the paper.

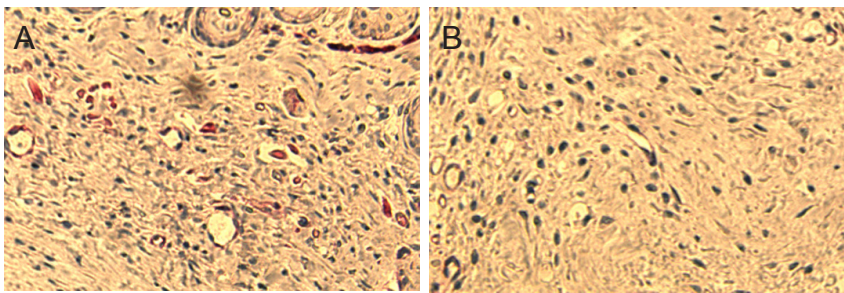
**Conflicts of interest:** None declared.

## References

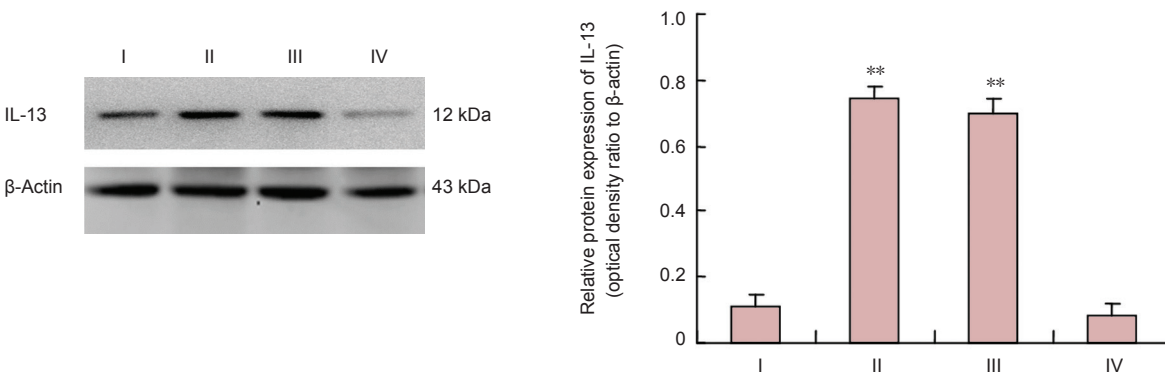
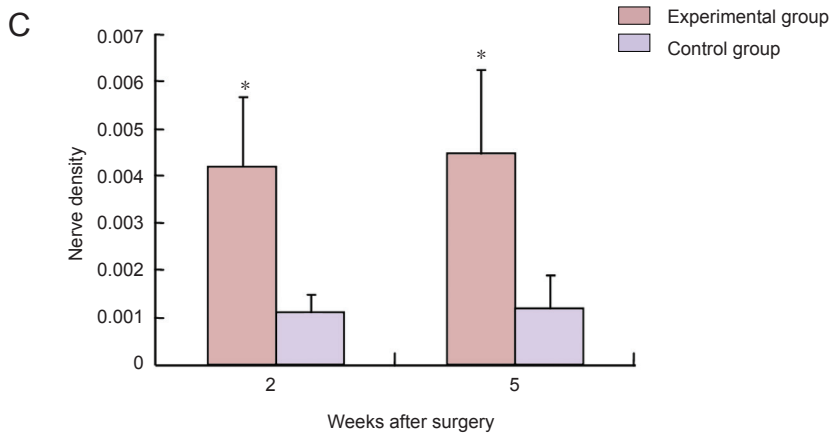
- Aarabi S, Bhatt KA, Shi Y, Paterno J, Chang EI, Loh SA, Holmes JW, Longaker MT, Yee H, Gurtner GC (2007) Mechanical load initiates hypertrophic scar formation through decreased cellular apoptosis. *FASEB J* 21:3250-3261.
- Akaishi S, Ogawa R, Hyakusoku H (2008) Keloid and hypertrophic scar: neurogenic inflammation hypotheses. *Med Hypotheses* 71:32-38.
- Akita S, Akino K, Hirano A (2013) Basic Fibroblast Growth Factor in Scarless Wound Healing. *Adv Wound Care* 2:44-49.
- Anderson JR, Fear MW, Phillips JK, Dawson LF, Wallace H, Wood FM, Rea SM (2011) A preliminary investigation of the reinnervation and return of sensory function in burn patients treated with INTEGRA®. *Burns* 37:1101-1108.
- Antony AK, Kong W, Lorenz HP (2010) Upregulation of neurodevelopmental genes during scarless healing. *Ann Plas Surg* 64:247-250.
- Atiyeh B, Janom HH (2014) Physical rehabilitation of pediatric burns. *Ann Burns Fire Disasters* 27:37-43.
- Barron L, Wynn TA (2011) Fibrosis is regulated by Th2 and Th17 responses and by dynamic interactions between fibroblasts and macrophages. *American journal of physiology Gastrointestinal and liver physiology* 300:G723-728.
- Bloemen MC, van der Veer WM, Ulrich MM, van Zuijlen PP, Niessen FB, Middelkoop E (2009) Prevention and curative management of hypertrophic scar formation. *Burns* 35:463-475.
- Buckley G, Metcalfe AD, Ferguson MW (2011) Peripheral nerve regeneration in the MRL/MpJ ear wound model. *J Anat* 218:163-172.
- Citak M, Backhaus M, Meindl R, Muhr G, Fehmer T (2010) Rare complication after VAC-therapy in the treatment of deep sore ulcers in a paraplegic patient. *Arch Orthop Trauma Surg* 130:1511-1514.
- Elsharawy MA, Naim M, Greish S (2012) Human CD34<sup>+</sup> stem cells promote healing of diabetic foot ulcers in rats. *Interact Cardiovasc Thorac Surg* 14:288-293.
- Feng YQ, Li X, Zhang R, Liu Y, Leng TT, Wang YB (2010) Remodeling of skin nerve fibers during burn wound healing. *Neural Regen Res* 5:1515-1520.
- Ferrante CJ, Leibovich SJ (2012) Regulation of Macrophage Polarization and Wound Healing. *Adv Wound Care (New Rochelle)* 1:10-16.
- Gaspar K, Kukova G, Bunemann E, Buhren BA, Sonkoly E, Szollosi AG, Muller A, Savinko T, Lauerma AI, Alenius H, Kemeny L, Dieu-Nosjean MC, Stander S, Fischer JW, Ruzicka T, Zlotnik A, Szegedi A, Homey B (2013) The chemokine receptor CCR3 participates in tissue remodeling during atopic skin inflammation. *J Dermatol Sci* 71:12-21.
- Hamed K, Giles N, Anderson J, Phillips JK, Dawson LF, Drummond P, Wallace H, Wood FM, Rea SM, Fear MW (2011) Changes in cutaneous innervation in patients with chronic pain after burns. *Burns* 37:631-637.
- Henderson J, Ferguson MW, Terenghi G (2011) The reinnervation and revascularization of wounds is temporarily altered after treatment with interleukin 10. *Wound Repair Regen* 19:268-273.



**Figure 1** Histological observation of scars in a mouse model of hypertrophic scarring (hematoxylin-eosin staining,  $\times 10$ ). (A) Mouse-loaded scars in the experimental group (formation of a hypertrophic scar by mechanical loading on the dorsum). (B) Control group developed very little fibrosis. Arrows show scars.



**Figure 2** Nerve fiber and nerve density in a mouse model of hypertrophic scarring. (A, B) Nerve fiber of skin (immunohistochemical staining,  $\times 40$ ). (A) Experimental group (formation of a hypertrophic scar by mechanical loading on the dorsum): brown S-100-immunoreactive nerve fibers, dot, short bar or bean sprout shapes. (B) Control group: no immunoreactivity of S-100 in scars. (C) Nerve density (nerve number/total dermal area) in scar tissues. Data are expressed as the mean  $\pm$  SEM. \* $P < 0.05$ , vs. control group (independent samples  $t$ -test).



**Figure 3** IL-13 expression in a mouse model of hypertrophic scarring (western blot assay). I: Control group at 2 weeks; II: experimental group (formation of a hypertrophic scar by mechanical loading on the dorsum) at 2 weeks; III: experimental group at 5 weeks; IV: control group at 5 weeks. Data are expressed as the mean  $\pm$  SEM. \*\* $P < 0.01$ , vs. control group (independent samples  $t$ -test). IL: Interleukin.

- Henderson J, Ferguson MW, Terenghi G (2012) The reinnervation pattern of wounds and scars after treatment with transforming growth factor  $\beta$  isoforms. *J Plast Reconstr Aesthet Surg* 65:e80-86.
- Hofmann U, Knorr S, Vogel B, Weirather J, Frey A, Ertl G, Frantz S (2014) Interleukin-13 deficiency aggravates healing and remodeling in male mice after experimental myocardial infarction. *Circ Heart Fail* 7:822-830.
- Hu MS, Rennert RC, McArdle A, Chung MT, Walmsley GG, Longaker MT, Lorenz HP (2014) The role of stem cells during scarless skin wound healing. *Adv Wound Care* 3:304-314.
- Ishiguro M, Ikeda K, Tomita K (2010) Effect of near-infrared light-emitting diodes on nerve regeneration. *J Orthop Sci* 15:233-239.
- Isoardo G, Stella M, Cocito D, Rizzo D, Migliaretti G, Cauda F, Palmiessa A, Faccani G, Ciaramitaro P (2012) Neuropathic pain in post-burn hypertrophic scars: A psychophysical and neurophysiological study. *Muscle Nerve* 45:883-890.
- Khattak MJ, Ahmad T, Rehman R, Umer M, Hasan SH, Ahmed M (2010) Muscle healing and nerve regeneration in a muscle contusion model in the rat. *J Bone Joint Surg Br* 92:894-899.
- Kim SY, Yang J, Lee YC (2011) The effects of nicergoline on corneal nerve regeneration in rat corneas after photorefractive keratectomy. *Curr Eye Res* 36:29-33.
- Kumar V (2014) Innate lymphoid cells: New paradigm in immunology of inflammation. *Immunol Lett* 157:23-37.
- Liu K, Gao Z, Wu X, Zhou G, Zhang WJ, Yang X, Liu W (2014) Knocking out Smad3 favors allogeneic mouse fetal skin development in adult wounds. *Wound Repair Regen* 22:265-271.
- Malin D, Sonnenberg-Riethmacher E, Guseva D, Wagener R, Aszodi A, Irintchev A, Riethmacher D (2009) The extracellular-matrix protein matrilin 2 participates in peripheral nerve regeneration. *J Cell Sci* 122:995-1004.
- Mammoto T, Seerattan RA, Paulson KD, Leonard CA, Bray RC, Salo PT (2008) Nerve growth factor improves ligament healing. *J Orthopaed Res* 26:957-964.
- Mandal D, Fu P, Levine AD (2010) REDOX regulation of IL-13 signaling in intestinal epithelial cells: usage of alternate pathways mediates distinct gene expression patterns. *Cell Signal* 22:1485-1494.
- Moore ML, Dewey WS, Richard RL (2009) Rehabilitation of the burned hand. *Hand Clin* 25:529-541.
- Morris MW, Allukian M, Herdrich BJ, Caskey RC, Zgheib C, Xu J, Dorsett-Martin W, Mitchell ME, Liechty KW (2014) Modulation of the inflammatory response by increasing fetal wound size or interleukin-10 overexpression determines wound phenotype and scar formation. *Wound Repair Regen* 22:406-414.
- Nakagaki O, Miyoshi H, Sawada T, Atsumi T, Kondo T, Atsumi T (2013) Epalrestat improves diabetic wound healing via increased expression of nerve growth factor. *Exp Clin Endocrinol Diabetes* 121:84-89.
- Ngeow WC (2010) Scar less: a review of methods of scar reduction at sites of peripheral nerve repair. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 109:357-366.
- Nguyen V, Conyers JM, Zhu D, Gibo DM, Dorsey JF, Debinski W, Mintz A (2011) IL-13 $\alpha$ 2-targeted therapy escapees: biologic and therapeutic implications. *Transl Oncol* 4:390-400.
- Niu Y, Miao M, Cao X, Song F, Ji X, Dong J, Lu S (2014) Infiltration of macrophages and their phenotype in the healing process of full-thickness wound in rat. *Zhonghua Shao Shang Za Zhi* 30:109-115.
- Novak ML, Koh TJ (2013) Macrophage phenotypes during tissue repair. *J Leukocyte Biol* 93:875-881.
- Oh MH, Oh SY, Yu J, Myers AC, Leonard WJ, Liu YJ, Zhu Z, Zheng T (2011) IL-13 induces skin fibrosis in atopic dermatitis by thymic stromal lymphopoietin. *J Immunol* 186:7232-7242.
- Park JS, Lee JH, Han CS, Chung DW, Kim GY (2011) Effect of hyaluronic acid-carboxymethylcellulose solution on perineural scar formation after sciatic nerve repair in rats. *Clin Orthop Surg* 3:315-324.
- Perry DM, McGrouther DA, Bayat A (2010) Current tools for noninvasive objective assessment of skin scars. *Plast Reconstr Surg* 126:912-923.
- Pradhan L, Nabzyk C, Andersen ND, LoGerfo FW, Veves A (2009) Inflammation and neuropeptides: the connection in diabetic wound healing. *Expert Rev Mol Med* 11:e2.
- Rashmi RN, Eckes B, Glöckner G, Groth M, Neumann S, Gloy J, Sellin L, Walz G, Schneider M, Karakesiosoglou I, Eichinger L, Noegel AA (2012) The nuclear envelope protein Nesprin-2 has roles in cell proliferation and differentiation during wound healing. *Nucleus* 3:172-186.
- Reichard M, Hovakimyan M, Guthoff RF, Stachs O (2014) In vivo visualisation of murine corneal nerve fibre regeneration in response to ciliary neurotrophic factor. *Exp Eye Res* 120:20-27.
- Romano MR, Biagioni F, Carrizzo A, Lorusso M, Spadaro A, Micelli Ferrari T, Vecchione C, Zurria M, Marrazzo G, Mascio G, Sacchetti B, Madonna M, Fornai F, Nicoletti F, Lograno MD (2014) Effects of vitamin B12 on the corneal nerve regeneration in rats. *Exp Eye Res* 120:109-117.
- Scott JR, Muangman P, Gibran NS (2007) Making sense of hypertrophic scar: a role for nerves. *Wound Repair Regen* 15 Suppl 1:S27-31.
- Semlali A, Jacques E, Koussih L, Gounni AS, Chakir J (2010) Thymic stromal lymphopoietin-induced human asthmatic airway epithelial cell proliferation through an IL-13-dependent pathway. *J Allergy Clin Immunol* 125:844-850.
- Shen L, Zeng W, Wu YX, Hou CL, Chen W, Yang MC, Li L, Zhang YF, Zhu CH (2013) Neurotrophin-3 accelerates wound healing in diabetic mice by promoting a paracrine response in mesenchymal stem cells. *Cell Transplant* 22:1011-1021.
- Soller EC, Tzeranis DS, Miu K, So PTC, Yannas IV (2012) Common features of optimal collagen scaffolds that disrupt wound contraction and enhance regeneration both in peripheral nerves and in skin. *Biomaterials* 33:4783-4791.
- Spits H, Di Santo JP (2011) The expanding family of innate lymphoid cells: regulators and effectors of immunity and tissue remodeling. *Nat Immunol* 12:21-27.
- Stein C, Küchler S (2013) Targeting inflammation and wound healing by opioids. *Trends Pharmacol Sci* 34:303-312.
- Stout RD (2010) Editorial: macrophage functional phenotypes: no alternatives in dermal wound healing? *J Leukocyte Biol* 87:19-21.
- Sun W, Lin H, Chen B, Zhao W, Zhao Y, Xiao Z, Dai J (2010) Collagen scaffolds loaded with collagen-binding NGF- $\beta$  accelerate ulcer healing. *J Biomed Mater Res A* 92:887-895.
- Tan J, Peng X, Luo G, Ma B, Cao C, He W, Yuan S, Li S, Wilkins JA, Wu J (2010) Investigating the role of P311 in the hypertrophic scar. *PLoS One* 5:e9995.
- Wang X, Zhao GQ (2010) Substance P and corneal wound healing. *Zhonghua Yan Ke Za Zhi* 46:857-860.
- Wang YB, Li X, Zhang R, Feng YQ, Liu Y (2010) Quantitative and morphological differences of nerve fibers between proliferative and mature scars in two- and three-dimensional spaces. *Neural Regen Res* 5:132-137.
- Weisser SB, McLaren KW, Kuroda E, Sly LM (2013) Generation and characterization of murine alternatively activated macrophages. *Methods Mol Biol* 946:225-239.
- Wise SK, Den Beste KA, Hoddeson EK, Parkos CA, Nusrat A (2013) Sinonasal epithelial wound resealing in an in vitro model: inhibition of wound closure with IL-4 exposure. *Int Forum Allergy Rhinol* 3:439-449.
- Wong VW, Paterno J, Sorkin M, Glotzbach JB, Levi K, Janusz M, Rustad KC, Longaker MT, Gurtner GC (2011) Mechanical force prolongs acute inflammation via T-cell-dependent pathways during scar formation. *FASEB J* 25:4498-4510.
- Yagmur C, Guneren E, Kefeli M, Ogawa R (2011) The effect of surgical denervation on prevention of excessive dermal scarring: A study on rabbit ear hypertrophic scar model. *J Plast Reconstr Aesthet Surg* 64:1359-1365.
- Yasui M, Shiraishi Y, Ozaki N, Hayashi K, Hori K, Ichianagi M, Sugiyama Y (2012) Nerve growth factor and associated nerve sprouting contribute to local mechanical hyperalgesia in a rat model of bone injury. *Eur J Pain* 16:953-965.
- Yates CC, Hebda P, Wells A (2012) Skin wound healing and scarring: fetal wounds and regenerative restitution. *Birth Defects Res C Embryo Today* 96:325-333.
- Zhang Z, Wang S, Tian X, Zhao Z, Zhang J, Lv D (2010) A new effective scaffold to facilitate peripheral nerve regeneration: Chitosan tube coated with maggot homogenate product. *Med Hypotheses* 74:12-14.