Wound Healing and Tissue Repair: A Trip Back to the Future

Chapter · January 2015

DOI: 10.1007/978-1-4939-2671-8_46

CITATION
1

READS
105

4 authors:

David G. Armstrong
University of Southern California
834 PUBLICATIONS **28,117** CITATIONS
SEE PROFILE

Nicholas Anthony Giovinco
The University of Arizona
27 PUBLICATIONS **253** CITATIONS
SEE PROFILE

Mahmoud A.Z. Abdelaal
The University of Arizona
1 PUBLICATION **1** CITATION
SEE PROFILE

Marvin J Slepian
The University of Arizona
205 PUBLICATIONS **3,059** CITATIONS
SEE PROFILE

Tissue Repair and Wound Healing: A Trip Back to the Future

46

Mahmoud A.Z. Abdelaal, Nicholas A. Giovinco, Marvin J. Slepian, and David G. Armstrong

Historical Introduction: From Healers to Doctors to Nurses and Teams

Early History Until the Early 1800s

Tissue repair and wound healing is one of the longest standing and central subjects in medicine and surgery. While records from antiquity date from the Pharaonic era (1500 BC), the basis of wound care likely had its origins much earlier extending to the period of prehistoric man where hunter-gatherers noticed that simple application of pressure to a bleeding wound and subsequent caring for its hygiene increased the chances of recovery from injury.

The finding of the Ebers Papyrus (Fig. 46.1), a document from ancient Egypt, provides insight into early Egyptian approach to wound care. It outlined the use of lint, animal grease, and honey as topical treatment of wounds. Lint provided a fibrous base that promoted wound-site closure, with the animal grease serving as a barrier to environmental pathogens, and honey functioning as an anti-infective agent [1]. The Greeks promoted similar therapies, but extended our insight into wounds to include characteristics of acute and chronic subtypes.

Claudius Galenus (better known as Galen of Pergamon) was a prominent Greek physician and a surgeon who furthered the understanding of wounds [2]. Significantly, he identified the importance of maintaining wound-site moisture to ensure successful closure of the wound.

In the twelfth century Moses Maimonides in his "Surgical Aphorisms" further identified the danger of skin "boils" and possible spread from these lesions, a precursor to the issue of infectious contamination and lateral spread. Further he identified the role of cauterization, heat, and corrosive agents as means of sterilizing a wound and preventing infectious spread [3].

All of these early observations and insight were amplified in the nineteenth century with the growth of the fields of microbiology and cellular pathology. See Fig. 46.2a–d. Finally, infection was recognized as being due to microorganism spread rather than a confluence of bad humors. Further, the role of inflammation, tissue ischemia, necrosis, and response to injury were revealed. These established the basis for a mechanistic understanding and approach to therapy.

M.A.Z. Abdelaal, M.D. • N.A. Giovinco, D.P.M. • D.G. Armstrong, D.P.M., M.D., Ph.D. (✉)
Southern Arizona Limb Salvage Alliance (SALSA), Department of Surgery, University of Arizona College of Medicine, Tucson, AZ, USA
e-mail: Dr.gm@live.com; ngiovinco@gmail.com; Armstrong@usa.net

M.J. Slepian, M.D.
Department of Medicine, University of Arizona College of Medicine, Tucson, AZ, USA
e-mail: chairman.syns@gmail.com

R. Latifi et al. (eds.), *Technological Advances in Surgery, Trauma and Critical Care*,
DOI 10.1007/978-1-4939-2671-8_46

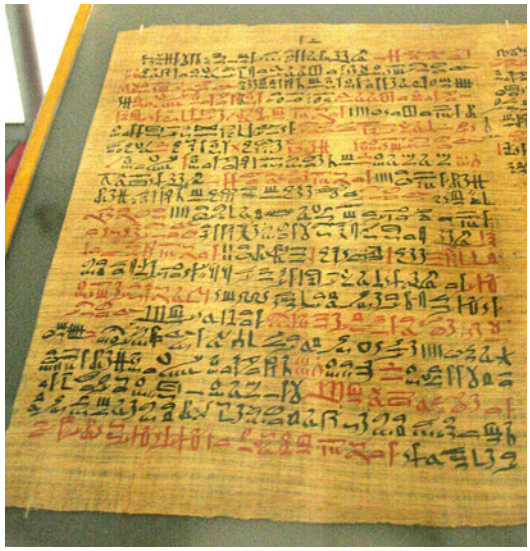

Fig. 46.1 The Ebers Papyrus (c. 1550 BC) from Ancient Egypt (*Source*: http://commons.wikimedia.org/wiki/File:PEbers_c41.jpg). By Einsamer Schütze [GFDL (http://www.gnu.org/copyleft/fdl.html) or CC BY-SA 3.0 (http://creativecommons.org/licenses/by-sa/3.0)], via Wikimedia Commons

Unfortunately, however, the same insights into the germ theory of medicine also led to physicians and surgeons literally and figuratively "washing their hands" off the problem. This perception coupled with the rise of professional societies of nurses led to much of the day-to-day management of wounds being borne on the shoulders of nurses.

1850s–1995: Poultices, Plasters, and Polymers

Much of the efforts over this period of time were focused on serving the dressing needs of nurses. Dressings were designed at best to manage drainage and to reduce pain during dressing changes. During World War I, in response to the needs of soldiers on the Western Front, Nobel Laureate Alex Carrel and chemist Henry Dakin screened more than 200 compounds and ultimately developed a wound-irrigating solution composed of sodium hypochlorite (0.45–0.5 %) in a buffered solution. This formulation, subsequently referred to as "Dakin's solution," provided excellent antimicrobial activity, combined with good irrigating properties without significant caustic, corrosive, or painful effects [4].

With the advent of synthetics and polymers in the 1950s, materials such as polyethylene, polypropylene, and polyvinyls provided constructs, in the form of gauzes and dressings, that further provided means to modulate the material properties of dressings. Some of these led to the first widely available foam/hydrocolloid-based dressings in the 1980s. More dressings such as these continued through the 1980s and into the 1990s until the next major advance: active healing technologies.

From Analog to Digital: Dressings and Supportive Care Meet Pharmaceuticals and Devices (Fig. 46.3)

In 1995, the first pharmaceutical requiring a prescription, Becaplermin (Regranex®, Smith+Nephew, Largo, FL), became available in North America. This single event led to rapid reeducation of the broader community of physicians and surgeons into chronic tissue repair and wound healing. Followed rapidly by negative pressure wound therapy (NPWT), and cell-based allografts (Dermagraft (Advanced Biohealing Ltd, La Jolla, CA), Apligraf® (Organogenesis, Inc., Canton, MA)), there emerged choices for physicians, surgeons, and nurses in therapies beyond previously available gauzes and intraoperative management/wound packing. In essence, technology further facilitated team building. This led to a dichotomy of sorts. The first of these two historical categories includes dressings and supportive care, which can now serve to manage drainage, reduce pain and frequency of dressing changes, and possibly also reduce/modulate bacterial load. The second, pharmaceuticals and devices, serves to promote angiogenesis and regeneration.

Significance of Wound Healing

Wound healing is a process via which injured tissue attempts to repair itself after injury. It is an intricate process in which the body tries to wall

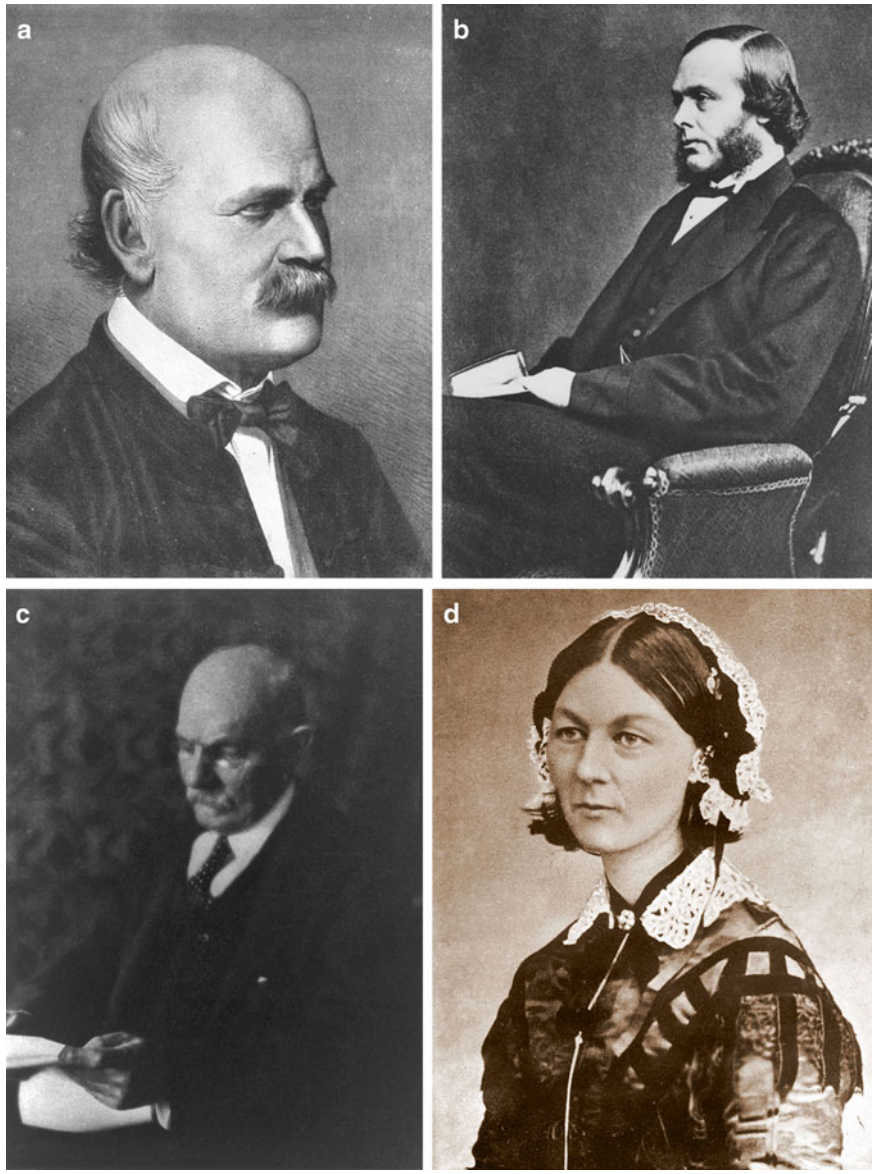

Fig. 46.2 (**a**–**d**) A brief timeline of wound healing. (**a**) Ignaz Philipp Semmelweis (July 1, 1818, to August 13, 1865) was a Hungarian physician now known as an early pioneer of antiseptic procedures (Ignaz Semmelweis 1860. Copper plate engraving by Jenő Doby. *Source*: http://upload.wikimedia.org/wikipedia/commons/f/f8/Ignaz_Semmelweis_1860.jpg. By Jenő Doby [Public domain], via Wikimedia Commons.); (**b**) Joseph Lister, 1st Baron Lister (April 5, 1827, to February 10, 1912). Known as Sir Joseph Lister. Between 1883 and 1897 was a British surgeon and a pioneer of antiseptic surgery (Joseph Lister, 1st Baron Lister, circa 1876: *Source*: http://commons.wikimedia.org/wiki/File%3AJoseph_Lister_c1867.jpg. Author unknown [Public domain], via Wikimedia Commons); (**c**) William Stewart Halsted (September 23, 1852 to September 7, 1922) was an American surgeon who emphasized strict aseptic technique during surgical procedures (William Stewart Halsted, 1852–1922, half-length portrait. *Source*: http://commons.wikimedia.org/wiki/File%3AWilliamHalsted.jpg. Author unknown [Public domain], via Wikimedia Commons); (**d**) Florence Nightingale (12 May 1820 to 13 August 1910) was a celebrated English social reformer and statistician, and the founder of modern nursing (Florence Nightingale from Care de Visite, circa 1850s. *Source*: http://commons.wikimedia.org/wiki/File%3AFlorence_Nightingale_CDV_by_H_Lenthall.jpg. By H. Lenthall, London [Public domain], via Wikimedia Commons)

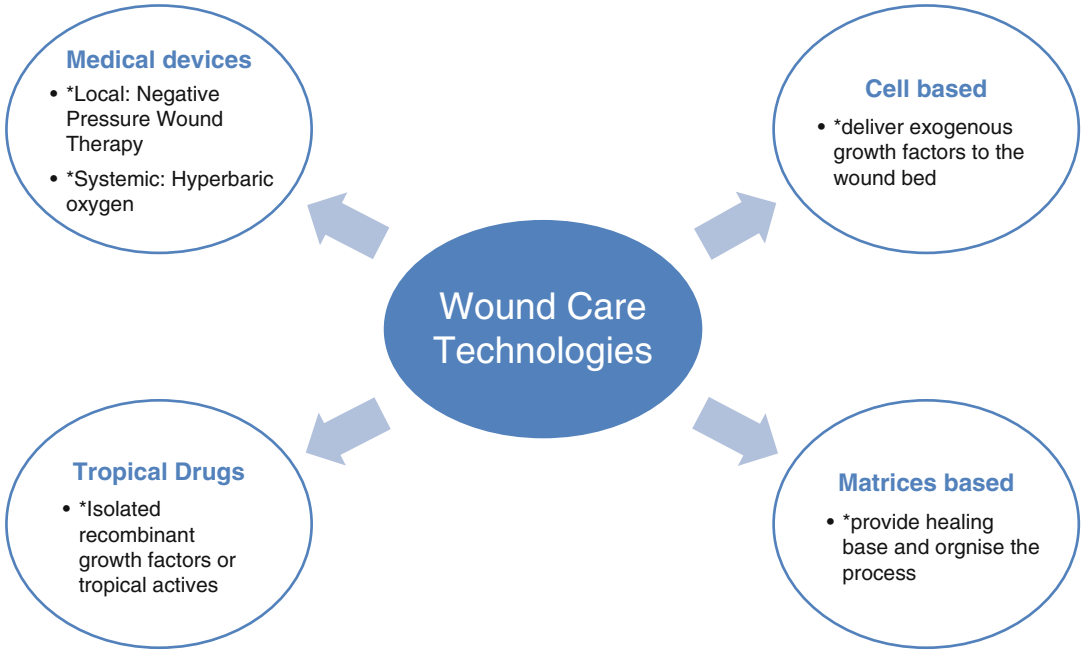

Fig. 46.3 Wound care technologies

off and eliminate infection, clear damaged and necrotic tissue, and rebuild damaged or lost tissue elements. The classic model of wound healing is divided into four sequential yet overlapping phases. These include (a) hemostasis, (b) inflammation, (c) proliferation, and (d) remodeling [5]. Under ideal conditions these phases progress synchronously. Certain pathophysiologic and metabolic conditions can alter this course of events so that healing is impaired or delayed, resulting in chronicity, i.e., chronic wounding [5, 6].

Perhaps the most common chronic wound in the developed and the developing world comes as a result of or concomitant with diabetes mellitus. According to the World health Organization, the epidemic of diabetes is affecting 285 million people (6.4 %) around the globe, and is expected to affect over 400 million adults by 2030 [7, 8]. A major cause of hospitalization and amputation among persons with diabetes is foot ulceration. It is estimated that one in four persons with diabetes in the USA will get foot ulcers at some point in their lifetimes [9]. As such, diabetic foot ulcer treatment is consuming nearly 25 % of overall cost of diabetes care, totaling around $43.5 billion [10]. Additionally, diabetes associated with ulceration increases the crude mortality rate from 6.7 to 27 % and shortens life expectancy by 3 years, relative to the average life expectancy for diabetic patients [11].

Additionally, as the world population is aging and becoming more sedentary, with an increased dependence on computers and automated manufacturing, low overall mobility results in venous obstruction or venous valvular dysfunction. This often precipitates chronic venous insufficiency which serves as yet an additional risk for foot ulceration. The annual prevalence of venous leg ulcers is estimated to be between 1.65 and 1.74 % in adults ≥65 years old, with anticipation of a substantial increase within the next several decades [12].

Wound Classification

In modern clinical practice, patients with foot ulcers present with a broad spectrum of underlying factors which contribute in the prognosis of the wound. Hence developing methods for stratification of wounds will help guide treatment decisions including the decision for amputation.

A recent advance was provided by the Society of Vascular Surgeons' Wound Ischemia Foot Infection (WIFI) classification, which was introduced in late 2013 to fill in the gaps remaining with other clinical diagnostic schemes. This classification systematically addressed individual grading of wound severity, ischemia, and presence of foot infection. Each category is graded based on specific criteria, which offers an improved and objective assessment of diabetic foot complications, and its recoverability [13].

The Strategies for Treatment: "Vertical" and "Horizontal"

There are two main strategies for treatment of ulcers, the "vertical strategy" or the "horizontal horizontal." In the vertical, the focus is aimed at trying to resolve the issue of the depth of the wound—either by filling the gap with living cells or approximating the edges using negative pressure. In the horizontal plan, the emphasis is aimed at trying to re-epithelialize the area using different techniques such as growth factors, negative pressure, stem cells, or amniotic membrane derivatives.

Modern Cell-Based Matrices: Three Examples

Grafix® (Osiris Therapeutics, Columbia, MD)

This is an example of a three-dimensional cellular matrix, designed to be directly applied to acute and chronic wounds—resulting from either burns or complications of the diabetic foot. It is a processed form of human placental membrane. With flexibility, this conforming membrane provides an excellent source of living mesenchymal stem cells (MSCs) and growth factors to the wound bed. The manufacturing process maintains the integrity of the extracellular matrix, the viability of the neonatal MSCs, and the active growth factors.

A clinical trial [14] to evaluate the safety and efficacy of Grafix for treatment of chronic foot ulcers was conducted. The trial included diabetic (both types) participants with foot ulcers which presented between 4 and 52 weeks, located around the malleoli aspects, varying in size from 1 to 15 cm^2, and age range from 18 to 80. The study protocol excluded patients with HbA1c above 12 %, active infection, osteomyelitis, cellulitis, or muscle, tendon, or joint capsule infections. Additional criteria were used to omit participants with circulatory insufficiency, which was determined using the ankle brachial index (ABI), toe brachial index (TBI), or Doppler. Participants with wound healing rates of 30 % or more during the screening period were also disqualified.

One hundred and thirty-nine patients were recruited, of which 42 were disqualified for matching the exclusion criteria. The remaining 97 were divided into two arms, 50 who received Grafix, with 47 receiving the standard wound therapy. No significant differences were observed in the baseline characteristics between the two arms. The Grafix arm revealed significant efficacy for primary and secondary endpoints when compared with the standard arm.

Thirty-one participants (62 %) of the Grafix group achieved complete wound closure, with significantly ($p=0.019$) faster median time (42 days) and probability (67.1 %) of closure, compared to ten in the standard wound therapy (21.3 %), with median time of 69.5 days and probability of 27.1 %. Grafix patients also required fewer study visits (i.e., applications) to achieve closure compared with patients in the control arm. The wound recurrence in the Grafix group, after complete closure, was 12.1 % less than the control arm.

As to safety, compared with the control arm, participants in Grafix arm experienced 22 % less adverse events, 18.2 % less wound-related infection, and 9 % fewer hospitalizations related to infection.

EpiFix® (MiMedx Group, Marietta, GA)

This is a dehydrated human amnion/chorion membrane (dHACM) allograft composed of multiple layers including a single layer of epithelial

cells, a basement membrane, and an avascular connective tissue matrix. EpiFix® is a minimally manipulated, dehydrated, nonviable cellular amniotic membrane allograft that preserves and delivers multiple extracellular matrix proteins, growth factors, cytokines, and other specialty proteins present in amniotic tissue to help regenerate soft tissue.

A clinical trial with similar enrollment criteria to the Grafix study outlined above was conducted to evaluate the efficacy of the product. However, the exclusion criteria were more restrictive. The study omitted patients with active Charcot arthropathy, wounds involving bone, or patients receiving immune system modulators.

The study enrolled 25 participants following the previous criteria; they were randomized and categorized into two groups, EpiFix ($n=13$) and standard wound care group ($n=12$). At the 4-week point, the wound size of the EpiFix group showed threefold size reduction relative to the standard group, and at the 6-week mark, 92 % of all ulcers in the Epifix group were completely healed compared to the control group (8 %). The mean time for wound closure in the EpiFix group was 50 % less than the standard.

During the study, only one patient in the Epifix group experienced an adverse event, while four participants in the control arm encountered side effects [15].

Dermagraft® (Advanced Biohealing Ltd, La Jolla, CA): Human Fibroblast-Derived Dermal Substitute

Human fibroblast-derived dermal substitute (Dermagraft, Advanced Biohealing Ltd, La Jolla, CA) is composed of fibroblasts, extracellular matrix, and a bioabsorbable scaffold. It is produced by culturing human dermal fibroblast cells derived from newborn foreskin tissue onto a bioabsorbable polyglactin mesh scaffold. As the fibroblasts proliferate filling the interstices of this scaffold, they secrete human dermal collagen, matrix proteins, growth factors, glycosaminoglycans, and cytokines to generate a three-dimensional, allogeneic, human dermal substitute containing metabolically active, living cells with a preferred, nearly parallel alignment of the collagen fibers within human dermal substitute [16]. Unlike human skin, human fibroblast-derived dermal substitute does not contain macrophages, lymphocytes, blood vessels, or hair follicles [17]. Studies suggest that this dermal substitute encourage healing via two mechanisms. Initially, Dermagraft offers living human dermal fibroblasts that deposit matrix proteins which facilitate angiogenesis. Secondly the construct delivers a preformed collagen matrix, receptors, and bound growth factors that help the migration and colonization of the host's epithelial cells, which promote wound closure [18, 19].

Greatest efficacy with this therapy was seen in the treatment of ulcers of greater than 6-week duration, suggesting that these ulcers are deficient in many of the factors necessary for healing [20]. In a study examining risk factors related to ulcer healing it was found that initial ulcers greater than 2 cm^2 were associated with a 150 % greater incidence of closure, while females were twice likely to heal more than males [21]. However, an episode of infection during the 12 weeks of treatment was associated with 3.4 times increased risk of non-closure cases.

A precaution that should be taken with Dermagraft relates to patients with known hypersensitivity to bovine products, as the packaging medium contains traces of bovine serum. Additionally the fibroblast-derived dermal substitute cannot be used in cases of clinical infection or ulcers with sinus tracts.

Devices: Negative Pressure Wound Therapy (Fig. 46.4a–d)

The main principle of the NPWT involves the application of local subatmospheric pressure to a defect, leading to a decrease of the wound margins and promotion of granulation tissue through enhancement of the surrounding circulation within the wound bed. Continuous negative pressure stimulates cell proliferation and increases extracellular fluid, which maintain moisture

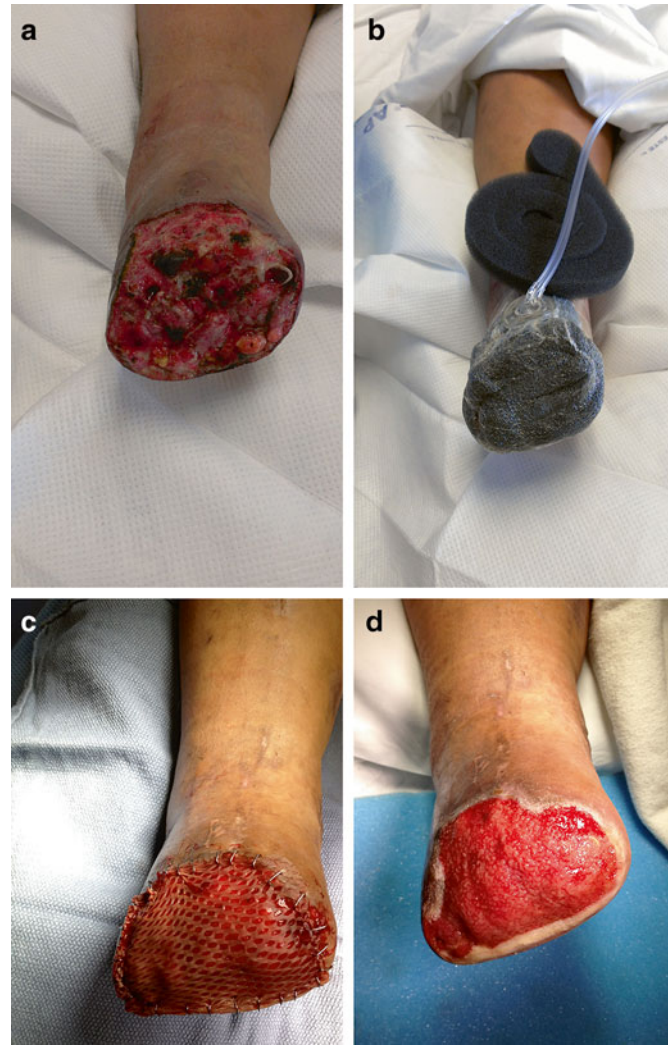

Fig. 46.4 (**a**–**d**) Negative pressure wound therapy (NPWT)

within the wound, as well as stimulate fibroblast migration and angiogenesis. Additionally, the system facilitates the removal of exudate and infectious material, and decreases matrix metalloproteinases (MMPs) which degrade tissue healing and block the surrounding microvascular circulation [22–27].

An early study of NPWT was conducted by Morykwas and colleagues [28], which reported a 400 % increase in the subcutaneous blood flow for tissue and muscles of Chester pigs. While they also observed a decrease of local blood flow after 5–7 min of the continuous negative pressure, they were able to reestablish high blood flow after a 2-min period of negative pressure.

Presently, there are several devices that allow for safe and effective NPWT. The most widely used system is the vacuum-assisted closure device, or VAC® (KCI, San Antonio, TX) therapy. The VAC device is offered in several forms, the black traditional and the more common, silver, and white foam. Traditionally, black GranuFoam™ (KCI, San Antonio, TX) is made of porous (400–600 mm) relatively hydrophobic,

polyurethane ether. However, VAC Silver has micro-bonded metallic silver impregnated into the foam itself, whereas the WhiteFoam version has a premoistened, hydrophilic foam made from polyvinyl alcohol. This form is effective especially when used as bolster for skin grafts and when granulation tissue formation is not the desired result, as in deep-space infection.

A study in 2005 by Armstrong and Lavery [29] over a 16-week period showed that the treatment with NPWT resulted in both an increased rate and proportion of patients healing complex lower extremity diabetic wounds following partial foot amputation. An additional study in 2008, by Blume and colleagues [30], compared NPWT with advanced moist wound therapy. Similarly this study demonstrated that NPWT was more effective with 14 % more complete closure observed.

The Future: Measuring What We Manage

While many new technologies show great promise in facilitating robust angiogenic response or promoting epithelialization, tissue repair and chronic wound healing have suffered from a lack of objective criteria for success. The only hard endpoint that presently exists (healing) has become inadequate to objectively assess the potential therapeutic effects of specific agents. Point of care and near-point-of-care diagnostics such as those that can assess protease activity, pH, cell receptor vitality, and general/specific organization of the wound microbiome will all likely play a crucial role in the development of new classes of therapeutics over the coming decade [31–33].

Conclusion

Stemming from antiquity, basic aspects of wounding and wound healing—both as to cause, pathophysiologic mechanisms, response to injury, and approaches to therapy—have been revealed and increasingly appreciated. In the modern era the role of ischemia, tissue injury, release of metalloproteases, inflammation, and infection are well recognized. Further, the time course of events is increasingly appreciated and therapeutic efforts aimed at these have been developed. Significant advances have been made in all mechanistic aspects of wound treatment including the novel use of negative pressure, biomaterial substrates, and cell engraftment. In the future further advances will continue to add to both our diagnostic and therapeutic capabilities. The future remains bright for this area.

References

1. Molan PC. Honey as a topical antibacterial agent for treatment of infected wounds. Nurs Times. 2001; 49(7-8):96.
2. Peter Brain G. Galen on bloodletting: a study of the origins, development and validity of his opinions, with a translation of the three works. Cambridge: Cambridge University Press; 2009. p. 1.
3. Rosner F. The medical legacy of moses maimonides. 1st ed. Jersey City, NJ: Ktav Pub Inc; 1997.
4. Hartmann H. Classic articles in colonic and rectal surgery. New procedure for removal of cancers of the distal part of the pelvic colon. Dis Colon Rectum. 1984;27(4):273.
5. Stadelmann WK, Digenis AG, Tobin GR. Physiology and healing dynamics of chronic cutaneous wounds. Am J Surg. 1998;176(2A Suppl):26S–38.
6. Mustoe TA, O'Shaughnessy K, Kloeters O. Chronic wound pathogenesis and current treatment strategies: a unifying hypothesis. Plast Reconstr Surg. 2006; 117(7 Suppl):35S–41.
7. Guariguata L, Whiting D, Weil C, Unwin N. The International Diabetes Federation diabetes atlas methodology for estimating global and national prevalence of diabetes in adults. Diabetes Res Clin Pract. 2011;94(3):322–32.
8. The Centers for Disease Control and Prevention. Number of people with diabetes increases to 24 million. http://www.cdc.gov/media/pressrel/2008/r080624.htm. Accessed 6 Oct 2014.
9. Singh N, Armstrong DG, Lipsky BA. Preventing foot ulcers in patients with diabetes. JAMA. 2005; 293(2):217–28.
10. Kantor J, Margolis DJ. Treatment options for diabetic neuropathic foot ulcers: a cost-effectiveness analysis. Dermatol Surg. 2001;27(4):347–51.
11. Brownrigg JR, Griffin M, Hughes CO, Jones KG, Patel N, Thompson MM, Hinchliffe RJ. Influence of foot ulceration on cause-specific mortality in patients with diabetes mellitus. J Vasc Surg. 2014;60(4): 982–6. e3.

12. Charles CA, Tomic-Canic M, Vincek V, Nassiri M, Stojadinovic O, Eaglstein WH, Kirsner RS. A gene signature of nonhealing venous ulcers: potential diagnostic markers. J Am Acad Dermatol. 2008; 59(5):758–71.
13. Mills Sr JL, Conte MS, Armstrong DG, Pomposelli FB, Schanzer A, Sidawy AN, Andros G. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: risk stratification based on wound, ischemia, and foot infection (WIfI). J Vasc Surg. 2014;59(1):220–34.e2.
14. Lavery LA, Fulmer J, Shebetka KA, Regulski M, Vayser D, Fried D, Kashefsky H, Owings TM, Nadarajah J. The efficacy and safety of Grafix® for the treatment of chronic diabetic foot ulcers: results of a multi-centre, controlled, randomised, blinded, clinical trial. Int Wound J. 2014;11(5):554–60.
15. Zelen CM, Serena TE, Denoziere G, Fetterolf DE. A prospective randomised comparative parallel study of amniotic membrane wound graft in the management of diabetic foot ulcers. Int Wound J. 2013;10(5):502–7.
16. Marston WA. Dermagraft, a bioengineered human dermal equivalent for the treatment of chronic nonhealing diabetic foot ulcer. Expert Rev Med Devices. 2004;1(1):21–31.
17. Wu SC, Marston W, Armstrong DG. Wound care: the role of advanced wound-healing technologies. J Am Podiatr Med Assoc. 2010;100(5):385–94.
18. Mansbridge J, Liu K, Patch R, Symons K, Pinney E. Three-dimensional fibroblast culture implant for the treatment of diabetic foot ulcers: metabolic activity and therapeutic range. Tissue Eng. 1998;4(4):403–14.
19. Mansbridge J. Skin substitutes to enhance wound healing. Expert Opin Investig Drugs. 1998;7(5):803–9.
20. Marston WA, Hanft J, Norwood P, Pollak R. The efficacy and safety of Dermagraft in improving the healing of chronic diabetic foot ulcers: results of a prospective randomized trial. Diabetes Care. 2003;26(6):1701–5.
21. Marston WA. Risk factors associated with healing chronic diabetic foot ulcers: the importance of hyperglycemia. Ostomy Wound Manage. 2006;52(3):26–8. 30, 32, passim.
22. Armstrong DG, Attinger CE, Boulton AJ, Frykberg RG, Kirsner RS, Lavery LA, Mills JL. Guidelines regarding negative wound therapy (NPWT) in the diabetic foot. Ostomy Wound Manage. 2004;50(4B Suppl):3S–27.
23. Dockery G, Crawford ME. Lower extremity soft tissue & cutaneous plastic surgery. 1st ed. Philadelphia: Saunders Ltd; 2006.
24. Andros G. Armstrong DG, Attinger C E, Boulton AJ, Frykberg R G, Joseph WS, Lavery LA. Morbach S, Niezgoda JA, Toursarkissian B. Consensus statement on negative pressure wound therapy (V.A.C. therapy) for the management of diabetic foot wounds. Ostomy Wound Manage 2006;Supp1:1–32.
25. Saxena V, Hwang CW, Huang S, Eichbaum Q, Ingber D, Orgill DP. Vacuum-assisted closure: microdeformations of wounds and cell proliferation. Plast Reconstr Surg. 2004;114(5):1086–96. Discussion 1097–8.
26. Olenius M, Dalsgaard CJ, Wickman M. Mitotic activity in expanded human skin. Plast Reconstr Surg. 1993;91(2):213–6.
27. Wysocki AB, Staiano-Coico L, Grinnell F. Wound fluid from chronic leg ulcers contains elevated levels of metalloproteinases MMP-2 and MMP-9. J Invest Dermatol. 1993;101(1):64–8.
28. Morykwas MJ, Argenta LC, Shelton-Brown EI, McGuirt W. Vacuum-assisted closure: a new method for wound control and treatment: animal studies and basic foundation. Ann Plast Surg. 1997;38(6): 553–62.
29. Armstrong DG, Lavery LA. Negative pressure wound therapy after partial diabetic foot amputation: a multicentre, randomised controlled trial. Lancet. 2005; 366(9498):1704–10.
30. Blume PA, Walters J, Payne W, Ayala J, Lantis J. Comparison of negative pressure wound therapy using vacuum-assisted closure with advanced moist wound therapy in the treatment of diabetic foot ulcers: a multicenter randomized controlled trial. Diabetes Care. 2008;31(4):631–6.
31. Isaac AL, Armstrong DG. Negative pressure wound therapy and other new therapies for diabetic foot ulceration: the current state of play. Med Clin North Am. 2013;97(5):899–909.
32. Armstrong DG, Giovinco NA. Diagnostics, theragnostics, and the personal health server: fundamental milestones in technology with revolutionary changes in diabetic foot and wound care to come. Foot Ankle Spec. 2011;4(1):54–60.
33. Fisher TK, Wolcott R, Wolk DM, Bharara M, Kimbriel HR, Armstrong DG. Diabetic foot infections: a need for innovative assessments. Int J Low Extrem Wounds. 2010;9(1):31–6.