Management of Complex Wounds with Fresh Cryopreserved Human Allografts

Adam Landsman, DPM, PhD

Abstract

In this manuscript, we review the use of fresh cryopreserved human skin allografts for the treatment of complex wounds. The reasons why this technology has generated renewed interest, as well as some clinical outcomes are examined. Based on our experience, we feel that cryopreserved human skin allografts offer an effective, relatively inexpensive alternative for the treatment of complex wounds such as diabetic foot ulcers and venous leg ulcers. *[N A J Med Sci. 2010;3(1):14-16.]*

Introduction

Management of complex wounds has seen great strides over the last 15 years. Arguably, one of the most significant milestones, FDA clearance of living skin equivalents, Apligraf and DermaGraft, opened the door to a wide variety of bioactive treatments designed to provide growth factors and living cells to the surface of a wound.[1,2] Prior to the availability of these two products, many surgeons relied on split thickness skin grafts, harvested from the patients' own body –usually the thigh or calf – to cover the wounds, and transpose growth factors and collagen to the wound site. However, this procedure frequently introduced new complexities including problems associated with the donor site such as pain, infection, and limited availability of appropriate tissue.[3] In addition, this would normally require a trip to the operating room for harvest. Because of the desire to have a split thickness graft, but the difficulties in procuring it, surgeons began to search for alternative tissue sources, and ultimately looked to tissue banks for human skin allografts. In this paper, we will look at various issues related to the use of human skin allografts, and review some cases where it is being used today.

Human Skin Allografts in the 1980's

The use of donated human tissues met a tremendous setback in the 1980's when people started to develop AIDS. Perhaps no modern disease was so widely feared and misunderstood as AIDS. Ambiguity about how the virus was spread, and the delay in the appearance of symptoms put everyone on guard. The idea of accepting tissues from an unknown source was widely discouraged, except in cases of most dire need.

Many companies opted to produce decellularized versions, in which the cellular components were removed from the complex collagen matrix while minimally altering its native collagen structure. Graftjacket (Wright Medical) was one of the earlier products available as a decellularized material, suitable for treatment of wounds. These products were safe, and could be packaged and stored for use outside of the operating room. Additionally, the removal of cellular components created a sense of safety with the public that there was less risk of virus transmission.

As many manufacturers entered the market with decellularized products made from bovine skin, porcine intestine, and equine pericardium, it became apparent that the decellualrization process could alter the structure of the collagen.[4] Intentionally induced cross-linking of the collagen polymer resulted in diminished incorporation due to resistance to enzymatic degradation. In some cases, heavily cross-linked decellularized grafts were rejected or encapsulated, rather than being incorporated into the wound bed.

Human skin is a complex composite made up of cellular components like fibroblasts and keratinocytes, and acellular components like collagen, which forms a scaffold for cellular infiltration, and can stabilize the surface of the wound. Tissue banks such as LifeNet Health (Virginia Beach, VA) began to develop safety procedures for screening, procurement, and storage several decades ago. These procedures continually improve the safety profile of these materials, and helped to reassure potential recipients of the safety of these grafts, and are federally monitored. To date, over 2 million tissue transplantations have occurred with LifeNet Health tissues, without a single incidence of infection of a recipient from donated tissue.

Fresh Human Allografts Make a Comeback

Living skin equivalents such as Apligraf and DermaGraft altered the way that many wound care professionals approached complex or chronic wounds. These materials provided a supply of living fibroblasts alone, or keratinocytes and fibroblasts, which released growth factors on contact, before they die. However, growth factors alone may not be

Adam Landsman, DPM, PhD
Beth Israel Deaconess Medical Center
Assistant Professor of Surgery
Harvard Medical School
Division of Podiatric Surgery
BIDMC
Baker 3, 1 Deaconess Road, Boston, MA 02215
Tel: 617-632-8428 Fax: 617-632-7090
Email: alandsma@bidmc.harvard.edu

sufficient. The collagen component is also necessary, and these materials were only provided in small quantities.

Whether using a collagen or a living cellular bioactive material, the gold standard has always been actual skin. Human allografts, harvested fresh, found wide usage for patients with burns. But delivery of fresh grafts presents a variety of other challenges, so fresh cryopreserved grafts have now emerged as the new gold standard for delivering biologically active materials to the wound bed.[5]

Clinical Experiences with Human Cryopreserved Skin Allografts

In our clinic, the two most common types of chronic wounds encountered are diabetic foot ulcers and venous stasis ulcers. Although their etiology is dramatically different, these chronic wounds have many factors in common. Many wounds are deficient in collagen and/or growth factors necessary to heal the wound. In some cases, other factors also may interfere in the healing process. Infections, poor blood supply, mechanical pressure, poor nutritional status, and chronically elevated glucose all may play a role in preventing wound closure regardless of what bioactive component is added to the surface. However, when these other factors have been optimized, the addition of bioactive components may provide the push necessary for a wound to achieve closure.

Fresh frozen human skin allografts (TheraSkin; LifeNet Health, Inc., Newport News, VA) are delivered to our site in a cryopreservative, packed on dry ice, and can be stored at -70^{o}F for months at a time. When needed, a quick thawing process is utilized. The package is opened, and room temperature normal saline is added to the packet to thaw the material and rinse away the cryopreservative. This rinsing process is repeated two more times, and the defrosted graft, complete with growth factors, and some living cellular components are now ready for application. Once defrosted, the skin can be stored in saline for several hours at room temperature.

Site preparation consists of debridement of the wound using a scalpel, leaving behind a clean wound, with little hyperkeratotic tissue or necrotic material on the surface (**Figure 1A**). Ideally, you will see bleeding at the site, but in wounds where profusion is sub-optimal, no bleeding may be apparent. The graft is retrieved from the saline solution and placed on the surface of the wound with the white (dermal) side facing the wound, and the pigmented (epidermal) side facing superficially (**Figure 1B**). Once in place, the graft can be stitched to the wound bed or simply held with several steri-strips (**Figure 1C**). We like to cover the graft with TheraGauze (Soluble Systems, Inc.; Virginia Beach, VA), a moisture regulating dressing which contains no petroleum products, and is also non-adherent. By regulating moisture on the wound surface, we avoid maceration while improving the chances that the TheraSkin will attach to the wound bed.

Human Allograft for the Treatment of Venous Leg Ulcers

In addition to local wound care, venous leg ulcers also require compression. We utilize Profore (Smith+Nephew), a 4 layered system of bandaging materials, designed to provide uniform compression for an extended period. This will help to reduce the venous fluid trapped in the leg, and helps reduce the internal pressure on the tissues.

Although compression is essential for achieving closure of venous ulcers, numerous studies have demonstrated that some wounds need more than just compression. Just like most chronic wounds, reapplication of growth factors and collagen frequently stimulates the healing process, and has become an important part of the therapy. Following debridement of the wound, the TheraSkin human allograft is applied, followed by the compression dressing. Once in place, the dressing and graft is left intact, and checked on a weekly basis. A review of 29 recent cases demonstrated that the average initial wound size was 4.84 cm2 (SD = 8.08cm^{2}), and required 67.4 days (SD = 50.6 days) and 2.47 grafts (SD = 1.93 grafts) to get the wound closed. A representative case is shown in **Figure 2**.

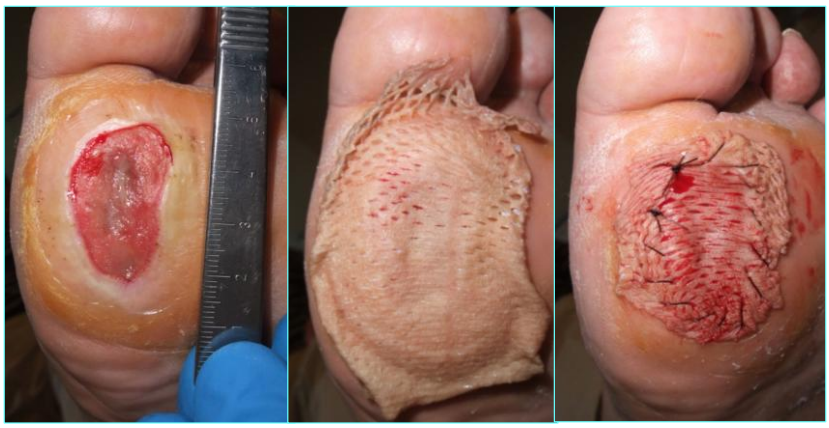


Figure 1. Application of TheraSkin cryopreserved skin allograft.
(1A) Wound is prepared by debriding the peri-wound callous and removing necrotic material from the wound base.
(1B) Following bed preparation, the TheraSkin is laid over the surface of the wound with the epidermal side facing superficial, and the demis (white) surface facing the wound bed.
(1C) The TheraSkin is shown stitched in place here. Alternatively, it can be held in place with steri-strips.

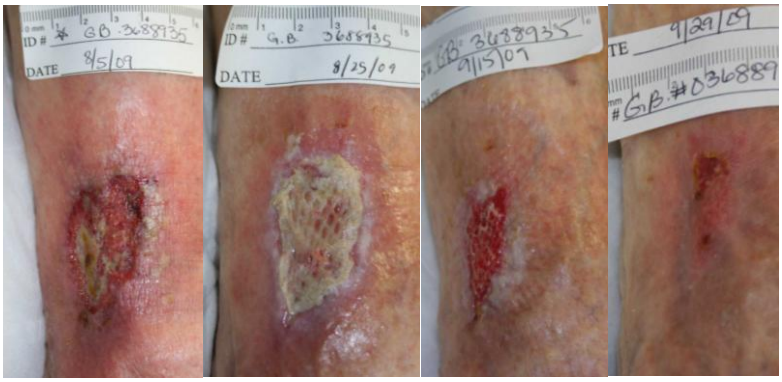
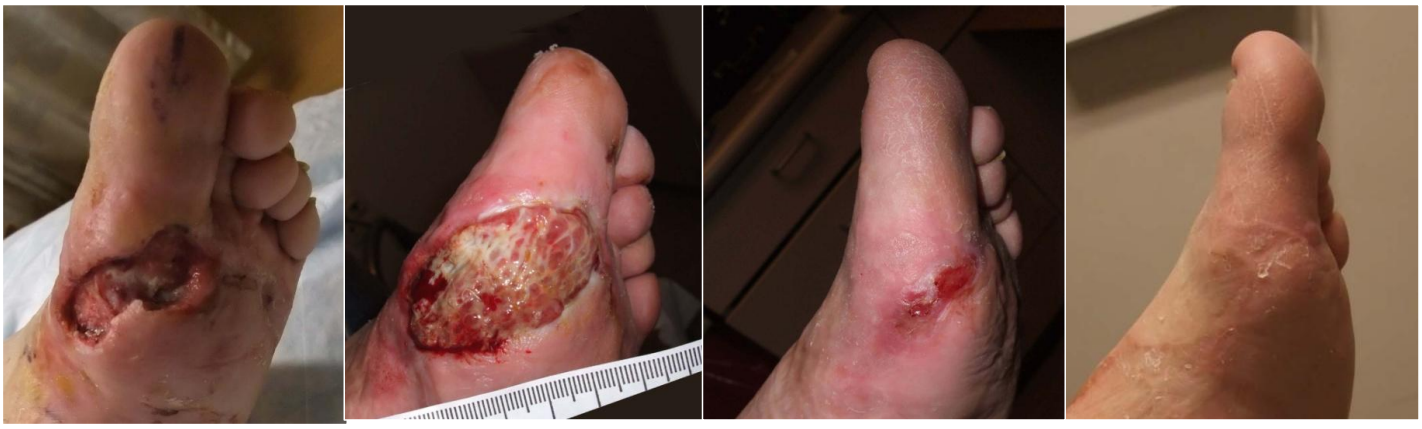


Figure 2. Venous Leg Ulcer. Initial wound was present for 11 months, and failed treatment with other advanced biologics. Once TheraSkin was applied, there is dramatic improvement, and the wound goes on to closure. (left)

Figure 3. This large diabetic foot ulcer was several months old, and was not showing any progress, despite standard wound care and off-loading. Originally, this wound measured 38cm^2. After preparation of the wound surface with negative pressure wound therapy, no decrease in size was noted. Subsequently, following local wound care and treatment with two TheraSkins, the wound closed in 19 weeks. (below)



Human Allograft for the Treatment of Diabetic Foot Ulcers

Just like venous leg ulcers, diabetic ulcers can be challenging to close, but their etiology is dramatically different from other types of chronic wounds. Diabetic foot ulcers are more likely to be associated with neuropathy, and repetitive mechanical stress. The condition is further exacerbated when glucose control is poorly regulated, vascular supply is diminished, and infection is present. As a result, they usually appear on the bottom of the foot. Therefore, in addition to local wound care, they require some steps to remove mechanical pressure from the wound surface. This can be accomplished with special shoe gear, casts, or even complete bed rest.

In our clinical practice, we utilize TheraSkin directly on the surface of the freshly debrided wound. After the graft is attached, the wound is covered with a moisture regulating, non-adherent dressing (TheraGauze), and left in place for five to seven days. We typically change the dressing on a weekly basis, and re-apply the graft every 2 weeks, as needed. A review of 14 recent cases demonstrated that the average initial wound size was 2.66 cm^2 (SD = 3.14cm^2), and required 49.3 days (SD = 26.3 days) and 1.93grafts (SD = 0.83 grafts) to get the wound closed. A representative case is shown in **Figure 3**.

Conclusion

In our clinic, we have had tremendous success with TheraSkin cryopreserved human allograft. It is readily available, relatively inexpensive, and supplies our chronic wounds with a variety of growth factors and collagen necessary to stimulate growth. TheraSkin has also been used successfully with a variety of other complex wounds, including pyogenic gangranosum, trauma related wounds, and post-operative dehiscence. As our ability to understand these complex wounds improves, I anticipate that additional benefits associated with human allograft wound treatment will become more apparent.

References

1. Veves A, Falanga V, Armstrong DG, Sabolinski ML; Apligraf Diabetic Foot Ulcer Study. A human skin equivalent, is effective in the management of noninfected neuropathic diabetic foot ulcers; a prospective randomized multicenter clinical trial. Diabetes Care. 2001;24(2):290-295.
2. Marston WA, Hanft J, Norwood P, Pollak R; Dermagraft Diabetic Foot Ulcer Study Group. 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):1701-1705.
3. Kirsner RS, Falanga V. Techniques of split-thickness skin grafting for lower extremity ulcerations. J Dermatol Surg Oncol. 1993;19(8):779-783.
4. Cornwell KG, Landsman A, James KS. Extracellular matrix biomaterials for soft tissue repair. Clin Podiatr Med Surg. 2009;26(4):507-523.
5. Spence RJ, Wong L. The enhancement of wound healing with human skin allograft. Surg Clinics North America. 1997;77(3):731-745.