Skin Expansion Technology in Acute Burns and Chronic Wounds

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ABSTRACT

The ability to grow skin has long been a topic of study and therapeutic interest. Currently, the main ways of doing this are 1) by placing tissue-expansion devices in the subcutaneous space and expanding skin over time, which can then be moved to cover contiguous structures, and 2) via processes that require relatively long (30 days) incubation periods to grow the patient's autogenous skin into laminar sheets. Over the past five years, there have been significant developments in the ability to expand skin cells, either at the bedside or in the laboratory, but much more rapidly than with previous methods. We explore and discuss the current skin cell-expansion techniques, focusing on point-of-care therapeutic interventions that can be used in the burn population as well as the chronic wound population, hair follicle stem-cell incubation techniques and studies supporting this therapy, as well as micro bullae grafting, and morcellated skin cell therapy. The current data supporting these therapeutic interventions and their current direction are outlined in detail.

INTRODUCTION

The technology of tissue expansion is gradually evolving as new innovative products are being developed to fill a dearth in optimal coverage options for acute burns and chronic wounds. The current gold standard remains the split-thickness skin graft (STSG), which, while dependable, has many disadvantages including donor-site morbidity, lack of a dermis, lack of skin appendages and eventual contraction and cosmesis issues. Placement of subcutaneous devices to expand local tissue for advancement has significant limitations. There has always been a need for robust skin-replacement therapies, as any number of causes—genetic disorders, cancers, chemical and thermal injury, trauma, diabetes, vascular disease, surgeries and more—can create difficult-to-heal wounds. These wounds and their treatment are a tremendous burden to patients and the United States healthcare system, with an annual cost of approximately $25 billion.[1] According to the National Institutes of Health, non-healing wounds affect over 5 million people in the United States, specifically individuals with disabilities, diabetes and the elderly. This number, along with the associated cost, is likely to continue to grow along with the prevalence of chronic diseases in our aging population.[2] Management of chronic wounds involves extensive office visits to wound care specialists, chronic wound care centers, and outpatient nursing, and often requires surgical interventions. Additionally, burns,

which can be a unique challenge in wound management due to the frequently extensive surface area involvement, which makes patients vulnerable to dehydration and opportunistic infections, as well as the need for numerous surgeries and a higher level of care with prolonged hospital stays, have always strained healthcare systems. The America Burn Association reports that nearly half a million burn injuries requiring medical attention occur annually in the United States and about 40,000 of those require hospitalization.[3,4] The World Health Organization estimates that about 11 million burn injuries occur annually worldwide, with a higher prevalence in low-income countries, which are resource-poor.[5] In high-income countries, the average annual healthcare cost per burn patient is estimated to be $88,000.[6] Unlike chronic wounds, burns are exceedingly prevalent in children, and account for 24% of burns in the U.S.,[3] which further expands the demographic requiring wound coverage and intensifies the need for a pliable functional skin coverage that can sweat and maintain its elasticity.

Human skin, our largest organ, is composed of three layers, the epidermis, dermis, and hypodermis, and each layer is composed of various cells that are synchronized to protect, regulate and renew. The multilayered epidermis, which contains keratinocytes, undergoes a constant cycle of desquamation and cell replacement. This self-renewal depends on stem cells located in the basal layers and is essential for wound healing. The strength of the epithelial tissue is thought to rely on the integrity of the vascularized and elastic dermis, which is rich in collagen and fibroblasts.[7] The hypodermis, which is composed mostly of adipose tissue, serves as a thermoregulator and barrier protecting underlying structures.[8] In general, both acute (burn) and chronic wounds are characterized and classified by their depth of injury. When any component of skin is damaged and the patient is predisposed to both local and systemic factors that influence or impede proper repair (poor tissue oxygen delivery, disrupted immune / inflammatory response, local bacterial metabolic strain, etc.), the process of wound healing can become dysregulated and lead to chronic wounds.

Full-thickness wounds that destroy all three layers necessitate re-epithelization from the edges of the wound, which can be near-impossible for larger wounds, especially without severe contracture and loss of functionality.[9] Furthermore, burn wounds, which regularly affect larger surface areas and at various depths, can trigger major fluid shifts, a cascade of caustic inflammatory markers, and an unprecedented hypermetabolic state-stresses on the body that can quickly turn to shock.[5] For severe burns, early wound closure reduces mortality, however, the time required for unaided reepithelialization is time not often afforded.[10] Without a skin barrier, the body cannot protect itself against certain external variables and is predisposed to infection.[11]

Although medical advancements in care for chronic wounds and burns, such as resuscitation, infection control, advanced wound dressings, and options for skin substitutes, have vastly improved outcomes, there is typically still a need for definitive tissue coverage. Restoration of the normal skin physiology is paramount to reduce infection, maintain elasticity, minimize contracture and reestablish the skin barrier. Unfortunately, it has been a challenge to achieve this with anything but human epithelial tissue. Surgical options generally include debridement of the wound and placement of split-thickness skin grafts, full-thickness skin grafts, or cellular or non-cellular biologic or synthetic products.[12] While STSGs, as already noted, are the gold standard of coverage options, this option is limited if there are insufficient areas of unburned skin to serve as a donor site.[13] Additionally, STSGs create a new and often painful wound. Skin substitutes show promise and function nicely as a bridge to definitive coverage, but are markedly expensive in addition to other limitations.[14] Whether the patient has acute burns or chronic wounds, speedy closure remains a challenge, and delay can be painfully problematic for physicians and patients alike. These wounds take a substantial amount of time to heal and require special care, attention, and resources, which explains the centuries of work leading up to more modern efforts to improve skin tissue engineering techniques, looking at which components of skin and what mode of delivery shows the most promise for wound coverage.

HISTORY OF SKIN EXPANSION

Just as fire, war, disease, and infections, which contribute to the vast majority of wounds, have plagued humanity for thousands of years, so have human efforts to heal these large skin defects. Healing these wounds has remained a significant medical and surgical challenge. Progress has been relatively stagnant given that the first known attempts at skin grafting were described by the surgeon Sushrutha in 600BC.[15] Tissue translocation and skin grafting were used to close facial wounds in India hundreds of years before awareness surfaced in the western world in the latter part of the 18th century.[16]

In 1817, Astley Cooper and Leroux des Tillets described skin graft techniques in Europe and Charles Buenger reported a full-thickness graft from the inner thigh to treat a nasal defect in 1821; these reports fueled interest in techniques to aid wound closure.[16] Innovations really took off following Jacques-Louis Reverdin's 1869 presentation at a meeting of the Société Impériale de Chirurgie de Paris showcasing "pinch grafts," which were piecemeal skin autografts for treatment of chronic venous ulcers.[17] Louis Ollier and Carl Thiersh are each credited for their successes with split-thickness grafts in 1872 and 1886, respectively.[18] The exact thickness was further investigated in 1929 by Blair and Brown, who suggested including deeper layers of dermis.[19] This increasing meticulousness in technique required tools with finesse; thus, surgical instruments underwent modifications as well. The double-bladed Catlin knife was replaced by the Thiersch knife, then the Humby knife, and so on throughout the turn of the century. Various modifications, such as protective guards and disposable razor blades, were made to improve the quality and consistency of skin grafts.[20,21] The invention of the dermatome by Earl Padgett in the 1930s permitted consistently even split-thickness grafts and remains the tool used today.[22] In 1908, Otto Lanz introduced the concept of meshing the graft for more surface coverage,[23] a practice that was further developed and used in 1964 by James Tanner, who touted its improved adaptability to irregular areas of the body, better drainage and thus better take.[24] The Meek technique, which was described in 1963, allowed the donor site-to-wound ratio to go from 1:3 to 1:9 by cutting the STSG into small, square tissue islands.[25] Although meshing and the Meek technique each significantly increased the surface area coverage ability without requiring an even larger donor-site wound, skin remains a finite resource, driving physician scientists to look for alternatives.

NON-SKIN EXPANSION OPTIONS: ALLOGRAFTS & XENOGRAFTS

Alternatives to autografts have been considered as sources of tissue coverage for a long time; reports on the use of both xenografts and allografts on human wounds date back to at least the 19th century. Initially, various creatures such as chickens, pigeons, cats, dogs, and cows were used in non-human xenografting experiments with limited success. Baronio attempted transplantation of skin between a cow and a horse,[26] while Dieffenbach attempted transplantation of pigskin to pigeons.[27] However, since these early investigators lacked knowledge of the basic principles of immunology, the disappointing outcomes were not surprising. The earliest record of a xenograft used, albeit temporarily, as skin replacement on humans with some success was in 1899 by Fowler, who used frogskin on large granulating wounds.[28] Since then, measures have been taken to find xenografts with low immunologic response, whether it be decellularized fish skin, fetal bovine dermis or geneticallly modified porcine skin, all of which can be used successfully as temporary wound coverage.[29] Similarly, efforts to lower the antigenicity of allografts or hamper the host's immune response have been made since the first attempts with cadaveric skin by Girdner in 1881.[30,31] Experiments with irradiation, attempts to inhibit immune triggering donor dendritic cells or host T cells, and trials with antioxidants or immunosuppressants to curb graft rejection have largely been ineffective.[32] The preservation of skin allografts with glycerol has been shown to reduce antigenicity, but only marginally prolongs graft take.[33] Even with rejection expected in about 10-14 days, skin allografts are still commonly used in burn centers as they remain a natural source of growth factors that promote healing and angiogenesis.[34] Fortunately, the inevitable rejection has not been found to hurt future uptake of an autograft and has been common practice prior to autografting.[35]

Modified allogenic skin grafts have been developed, eliminating the need to obtain a biopsy from the patient, minimizing antigenicity, and permitting advanced preparation. These skin substitutes replace either epidermis, dermis or both, and have varied compositions: allogenic, xenograft, or biosynthetic.[36] The commercially available products Dermagraft® and Apligraft® (Organogenesis, Canton, MA, USA) are derived from neonatal tissue and have both been approved for the treatment of non-healing diabetic foot ulcers. Dermagraft® is a cellular dermal substitute that includes fibroblasts and keratinocytes. Apligraft® is a composite allograft that consists of a layer of bovine collagen gel with neonatal fibroblasts acting as its dermis and an epidermal layer composed of neonatal keratinocytes.[36] These cultured keratinocyte allografts, which release numerous cytokines, are a potent stimulus for wound healing from the periphery, but do not appear to survive permanently in the wound bed.[37] Beele et al. reported a notable decrease in wound size in all but 2 of 16 non-healing leg wounds and complete closure in 62% at 8 weeks with the use of epidermal allografts.[38] Fratianne et al. reported faster healing of STSG donor sites with keratinocyte allografts than without.[39] Xenografts, allografts and combinations of the two are readily available and have all been proven to protect wounds, decrease bacterial count, minimize pain, and stimulate growth. However, despite all the scientific progress made in the last century, these options truly remain impermanent solutions that only better prepare the wound bed for a lasting autograft.

Many comprehensive reviews of the available literature on xenografts have been previously published and are outside the scope of this article's focus on autogenous skin-expansion therapy.

CULTURED EPITHELIAL AUTOGRAFTS

The first major breakthrough to address the limitations of autologous skin grafting was the creation of cultured epithelial autografts (CEAs) in 1975 by Rheinwald and Green.[40] CEAs are obtained from skin biopsies of the epidermal layer and stem cell keratinocytes are cultured in vitro to create epidermal-like tissue. The first clinical application was by O'Connor et al. in 1981 for two patients with 40-80% total body surface area burns who were treated with both CEAs and STSGs. The direct comparison showed no major differences in fragility or contraction.[41] In this technique, a small sample of uninjured skin is biopsied, usually at the axilla or pubis. Epidermal cells are isolated and plated on a layer of mesenchymal "feeder cells" which helps promote keratinocyte growth. After 3-4 weeks, the CEA sheets are 8-10 cell layers thick and can be enzymatically detached from the culture vessel and transplanted back onto the patient's wounds.[42] Initial clinical trials focused on the application of CEAs in burn patients.[43-45] During the 1980s, they were applied to other large skin defects, including pyoderma gangrenosum,[46] congenital nevi,[47] and chronic leg ulcers.[48]

CEAs have the benefit of not introducing a large secondary wound to cover the initial wound, unlike traditional skin-grafting techniques. Wound contraction is minimal, and this technique can be used in areas of the body with frequent mechanical stress, such as the eyelids, fingers, and toes.[43] However, there are several pitfalls, including near-month-long delays with culturing and obtaining the in vitro epithelial sheets and high costs associated with production, requiring a laboratory and specialized personnel. The reports of uptake vary, particularly for full-thickness burns,[49] and CEAs are considered to be less effective than traditional STSGs. A major complaint is the fragility and friability of the cultured sheets;[50,51] even after take, CEAs lack durability and can easily shear, blister or avulse for months after grafting. CEAs are particularly vulnerable to bacterial proteases and cytotoxins in the first few weeks of placement, and an infection can cause a complete lack of uptake of the graft.[52]

Given these limitations, modifications of CEA grafts have been developed in which preconfluent keratinocytes are transferred to the patient prior to developing into sheets, with confluence and differentiation occurring in vivo. The keratinocytes are cultured on a delivery membrane that is subsequently inverted and placed on the wound. Several different matrices have been used to culture keratinocytes; both biologic (collagen, hyaluronic acid, fibrin glue, and acellular porcine dermis) and synthetic (polyurethane, polymeric film, Teflon film, Poly(hydroxyethyl Methacrylate, Celltran, and spherical microcarriers).[52] Ronfard et al. looked at long-term outcomes of CEA transplantation when grown on fibrin matrix. In their study, a young female burn victim who required CEA transplantation to her abdomen was able to carry three successful pregnancies without complications, demonstrating this tissue's ability to withstand stretch and mechanical stress.[50]

Currently, very few CEAs are available commercially. Although CEA technology was developed before the FDA published

regulations on cell therapies, Epicel® (Vericel, Cambridge, MA, USA) received swift approval in 2007 under the Humanitarian Device Exemption for use in burns when the total burn surface area (TBSA) is greater than or equal to 30%. This CEA is a prepared sheet 2-8 cell layers thick that takes 16-21 days to prepare and may be used either alone or in conjunction with split-thickness autografts.[53] Hickerson et al. summarized the largest cohort of CEA-treated patients to date. This review considered a dataset spanning 1989 to 2015, and included 954 patients who were treated with CEAs for severe burns at mostly U.S. hospitals, as well as 4 hospitals outside of the U.S. Both adult and pediatric patients were included, and these patients had a mean TBSA of 67%; 72% of the patients required only one harvest, and were treated with a mean of 105 CEA applications with 68% graft take. Overall, when compared to patients in the National Burn Repository with comparable burns, mortality rates were lower for those treated with CEAs in addition to conventional STSGs for large burns.[54] While this renewability is important to note, CEAs are still limited by delayed availability and a precarious nature, which diminishes the product's economic value and explains why they are only truly indicated for a small subset of patients-severe burn patients with no real alternative therapeutic options.

Epidermal Bullae Grafting

Harkening back to Reverdin's pinch grafts, the CelluTome™ Epidermal Harvesting System (3M-KCI, St. Paul, MN, USA) is an epidermal skin-grafting device that is used to treat chronic wounds. Using both heat and suction on the donor site over 30-45 minutes, this device (Fig. 1) creates up to 128 epidermal microdomes or bullae that can be harvested and transferred directly to the wound with an adhesive (Fig. 2).

In a retrospective case series, this autologous suction blister epidermal grafting (SBEG) technique was applied to 22 patients with chronic lower-extremity wounds and the average reepithelization rate was 88% at 2.5 months.[55] Another case series that examined the use of SBEG in 13 patients with "stalled" chronic wounds had an average 63% healing rate; 4 of the 13 patients had completely healed at 1 month and 8 had healed by 4 months.[56] Although these studies are clearly limited by the lack of an appropriate control group and the fact that most of the patients were receiving adjunctive wound treatments as well as multilayered compressive dressings, the anecdotal results are still encouraging. There appears to be accelerated wound healing with little downside as long as the wound is superficial enough to only require epidermal grafting. This technique is an office-based procedure, does not require any anesthetics, and has minimal donor-site morbidity. As concluded in a systematic review and meta-analysis that included seven articles on the efficacy of epidermal grafting for wound healing, although a 70% healing rate was achieved in 209 wounds, a randomized control trial that compares the outcomes to those with STSG or conservative therapy is still necessary given the heterogeneity of these studies and the lack of controls.[57]

CELL SUSPENSIONS & SPRAY-ON PRODUCTS

Given the challenges faced using sheets of CEAs, epithelial cell suspensions, either cultured or non-cultured, have gained popularity due to their ease of application and reduced preparatory time. These techniques require less donor skin, are less finnicky and avoid the need for a laboratory (unless cultured), which means that suspensions can be prepared on the spot during the surgical operation. Hunyadi et al. reported the first use of non-cultured keratinocyte suspensions for the treatment of both burn wounds and chronic wounds in 1988. After patients' wounds were treated with a fibrin matrix either with or without keratinocytes, within 14-21 days, the group with added keratinocytes had healed completely compared to the group without. These initial findings suggested that added keratinocytes hastened closure of partial- and full-thickness wounds.[58] Migliano et al. saw good results using autologous non-cultured epidermal cell suspensions in combination with lipofilling for the reconstruction of laser-ablated facial wounds in skin cancer patients, suggesting it holds promise for correcting skin graft sequela and recapturing a more natural appearance.[59]

In the past 25 years, attention has also been focused on spray-on techniques for the application of suspended autologous

Figure 1. CelluTome™ Epidermal Harvesting System (3M-KCI, St. Paul, MN, USA).

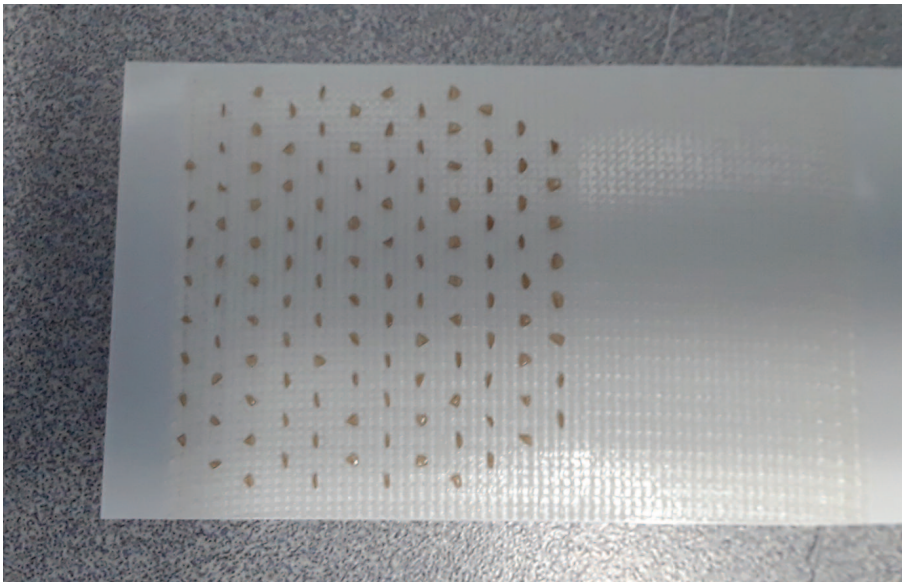

Figure 2. Harvested epidermal microdomes on adhesive.

keratinocytes. In 1998, Fraulin et al. first showed the potential of autotransplantation of epithelial cell suspensions delivered by an aerosolization apparatus in a pig model. They proved that aerosolized cells stayed viable, could be applied uniformly and could proliferate once laid down.[60] Shortly thereafter, Navarro et al. reported the use of a cell spray apparatus for the application of autologous keratinocytes in addition to split-thickness skin grafts in pig models.[61] In this study, pig models underwent full-thickness excision of 16 wounds; 8 of the wounds were subsequently treated by the application of 3:1 STSG and the spray-on application of a keratinocyte suspension and 8 of the wounds were treated with STSG alone. Overall, greater wound re-epithelization was seen in the keratinocyte suspension group at days 5 and 8 after grafting compared to the non-keratinocyte group (split-thickness skin grafting only), which promoted further interest in the spray-on technique.[61] The use of epithelial cell-spray for human wounds was championed by Dr. Fiona Wood, whose initial clinical studies corroborated the promising findings in animal studies showing the benefits of adding adjunct CEA treatment to conventional burn care.[62]

However, despite some of the initial promise with spray-on cell suspensions, a large prospective trial of non-autogenous cultured skin cells applied to venous leg ulcers (HP-802-247, Healthpoint Biotherapeutics, Fort Worth, TX, USA) failed to detect efficacy over a placebo group during phase 3 clinical trials.[63,64] As a result, additional trials with HP-802-247 were terminated early. The spray contained cryopreserved, growth-arrested fibroblasts and keratinocytes from neonatal foreskin delivered in a fibrin sealant-based matrix. It was theorized that batch-to-batch variability could have contributed to its failure. Yet, in a deeper review of enrolled subjects, it was clear that patient and wound variables, such as wound duration, bacterial species present, area and location, all greatly influenced healing.[65] Investment in this product and this trial contributed to the further development of the spray technique, with a focus on autologous cells rather than allogenic keratinocytes.

AUTOLOGOUS SKIN CELL SUSPENSION

ReCell® (Avita Medical, Cambridge, UK) is a point-of-care autologous skin cell suspension (ASCS) used in a spray-on fashion for the treatment of burns and chronic wounds. The technology behind ASCS was first introduced by Dr. Wood in 1993 under the trade name Spray-on-Skin™. For years, this product did not gain any traction in the medical world until after the 2002 Bali bombings, which put 28 severely burned patients in Dr. Wood's burn unit.[66] After successful treatment of these burns with spray-on skin cells, the product gained recognition and, in 2005, it was reintroduced to the market under the name ReCell®. The major advantage of this technique is that it permits for a smaller donor site and can be prepared in the operating room. The ASCS method makes it possible to minimize the harvest area by taking a small sample of the patient's skin (usually 1 cm^2 to cover an area of 80 cm^2) and then breaking down the harvested skin using the enzyme trypsin for 10 minutes. On the all-in-one device (Fig. 3) the epidermis is gently scraped off the dermis with a scalpel, and these cells are then suspended in a buffer of lactated Ringer's solution and filtered before this suspended cell solution is sprayed onto the wound bed (Fig. 4).

Numerous randomized trials from 2007-2018 showed that the use of ASCS was associated with a reduced time for the donor-site wound to re-epithelize, less scarring at the donor site, and reduced frequency of donor-site cellulitis.[67-69] It is currently FDA-approved for direct application to acute partial-thickness thermal burn wounds or for use in combination with meshed autografting for the treatment of all sizes of acute full-thickness burn wounds in adult patients.[70]

In one of the earliest clinical trials (in 2007), Gravante et al. compared the effectiveness of ASCS to standard STSG for the treatment of deep partial-thickness burns; 42 and 40 patients were enrolled in each group, respectively.[71] This research showed aesthetic and functional outcomes similar to those with the classic grafting technique, however, while the average areas of the burns treated were comparable between the two arms (176 cm^2 for ASCS vs 180 cm^2 for STSG), the average donor area harvested was notably smaller in the ASCS group (2.2 cm^2 versus 110 cm^2 for STSG).[71] These results were reproduced by a similar prospective study performed by Holmes et al., who compared ASCS to STSG to demonstrate their effectiveness on burns.[67] Of the 101 patients enrolled in that study, only 87 completed the full 52-week follow-up with no issue. The average percent area of burn was between 5.5 and 14.5. The patients who received ASCS showed healing similar to that with a split-thickness skin graft, but needed less harvest of skin grafts to cover a wider area.[67] Hayes et al. showed that ASCS can also be applied to chronic wounds. By week 14, patients treated with ASCS and compression had a significantly greater reduction in ulcer size

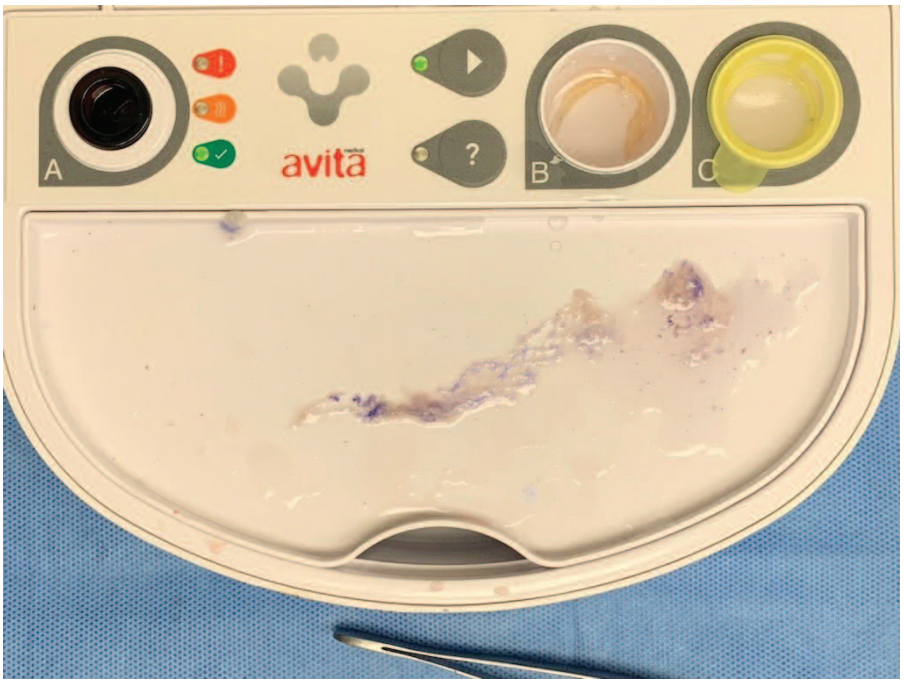

Figure 3. Preparation of ASCS on a sterile back-field table.

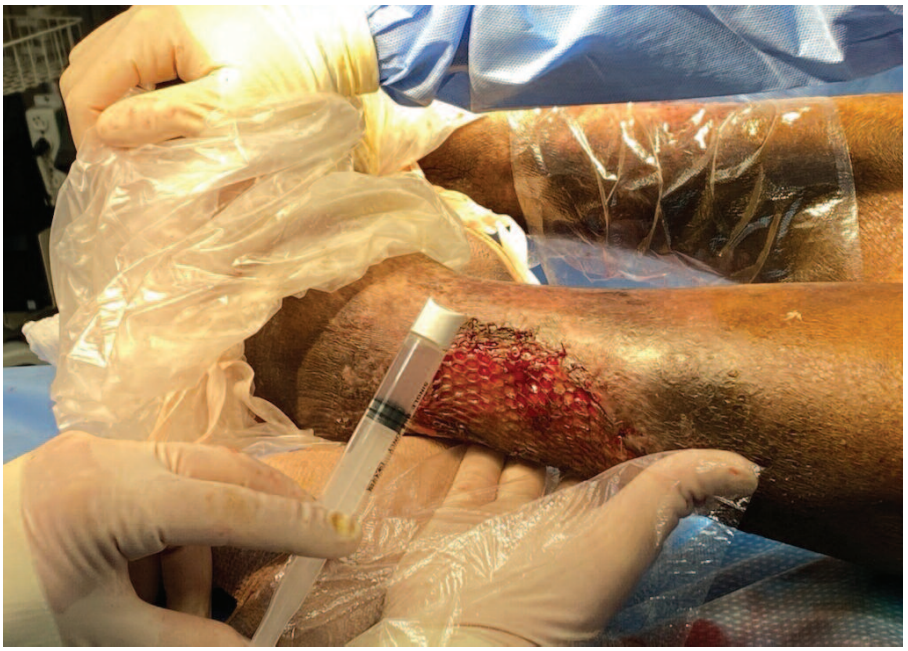

Figure 4. Application of ASCS on top of STSG.

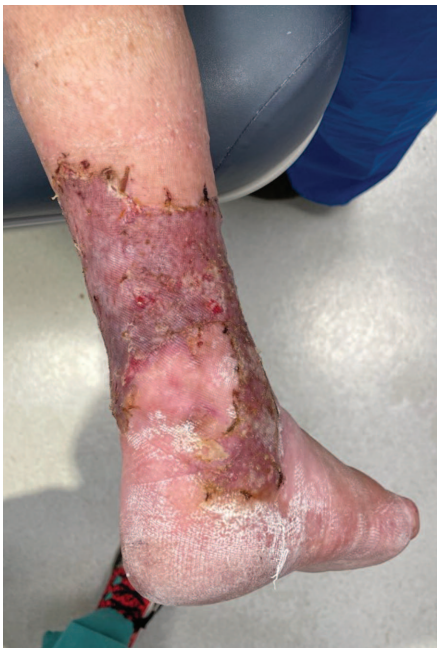

Figure 5. 2-week post-op visit after STSG and ASCS.

(8.94 cm^2 versus 1.23 cm^2, P = .0143), as well as less pain and better quality of life when compared to the control.[72] Anecdotally, we see more rapid filling of mesh interstices and complete reepithelialization of donor sites when ASCS is applied in conjunction with STSG in chronic wounds (Fig. 5). For surgical wounds, Wood et al. applied ASCS to a pediatric patient with a congenital melanocytic nevi lesion after dermabrasion and saw complete reepithelialization on day 8 and excellent pigmentation and texture at 5 months.[73]

In addition to non-cultured spray-on autologous cells, cultured epithelial autologous cells are available from the same company that offers ReCell® (Avita Medical). CellSpray® and CellSpray® XP (Avita Medical) are 5- to 7-day and 48-hour, respectively, harvested autologous cell suspensions that are processed in an external laboratory and applied with an aerosol system. Zweifel et al. found that, with the use of CellSpray® in three patients with full-thickness burns, wounds healed rapidly and there were no hypertrophic scars at 6 months.[74]

The CellMist™ Solution and SkinGun™ device (RenovaCare, Scottsdale, AZ, USA), which are still being developed, comprise another autologous skin spray system meant for the immediate treatment of wounds. After a biopsy is taken, the harvested skin including skin stem cells are processed briefly in the laboratory before application via the SkinGun™; the total process takes 90 minutes.[75] When used on six patients with partial-thickness burns, ASCS showed evidence of reepithelization by POD 3-6, and the patients were fully healed by 2 weeks.[76] Another study on the use of ASCS in 44 patients with deep partial-thickness burns helped to troubleshoot the technology for future clinical studies, and overall gave satisfying results in 75% of patients as an alternative to mesh autografting.[77]

MINCED SKIN GRAFTS

Similar to ASCS, minced skin (MS) grafting permits for transplantation of small autologous particles of skin onto wounds to accelerate reepithelization in a minimally invasive manner that could be performed in the office. The difference is that, for MS, the epidermis and dermis are not enzymatically separated, and the technique is more akin to a haphazard Meek technique. As mentioned above, the Meek technique involves dividing skin graft tissue into micrografts before application to expand the tissue nine-fold.[25] For MS, donor sites can be harvested under local anesthesia and are typically small full-thickness excisions from an unaffected area that are then chopped up and made into a paste or suspension by mixing in a saline or hydrogel-type solution. The grafts include portions of the dermis, epidermis, and skin appendages, so that diverse cell types, not just keratinocytes, are applied to wounds. Svensjö et al. tested the application of cultured keratinocytes, non-cultured keratinocytes, STSG and minced skin on 90 wounds in 3 different pigs. Wounds transplanted with MS and keratinocyte suspensions contained several colonies of keratinocytes at 2 weeks, but MS appeared to re-epithelialize faster than in non-cultured keratinocyte grafting. In addition, they could be prepared faster than the slightly better-performing cultured keratinocytes and had delayed contracture similar to STSG-treated wounds.[78] Miyanaga et al. performed a comparative study on 30 human subjects in whom they applied MS (taken from remaining STSG) to half of the STSG donor sites and saw both a significantly improved healing time and better cosmesis.[57] Advantages of MS are that donor sites are small, the graft can be harvested under local anesthesia, meaning it can be done in the office and quickly, and it provides progenitor cells along with keratinocytes making for faster healing. Similar to ASCS, MS can be used alone or in conjunction with STSG.

AUTOLOGOUS HETEROGENEOUS SKIN CONSTRUCT

The treatment of chronic wounds with autologous skin cells is also being studied by PolarityTE, a publicly traded company (Salt Lake City, UT, USA). They originally marketed their product SkinTE® as a minimally manipulated human tissue; during that period there was significant previous clinical experience,

with numerous positive publications. However, due to regulatory changes at the FDA, the company has moved to pursue approval via a Biologic License Application. This will have many positive effects with regard to the eventual availability of the product. Before these regulatory pathways, this product was referred to as an autologous homologous skin construct. However, to match the change in the regulatory environment, it is now referred to as an autologous heterogeneous skin construct (AHSC). In addition, the product is now being evaluated in a Phase III pre-market approval study (NCT 05372809) in Wagner 2 diabetic foot ulcers. AHSC is part of a 3-part process. First, clinicians harvest a full-thickness portion of skin (approximately 1 cm x 3 cm) from an unaffected area, and then send the sample to an FDA-registered facility for AHSC manufacturing, where in part the hair follicle pluripotent stem cells are isolated, and the viability of the substrate is assayed before being returned to the clinician for reapplication within 4 to 6 days. The material is returned in a paste form, which is delivered via syringe. (Fig. 6). Much of this technology is currently heavily protected intellectual property of the company. After the clinician debrides and prepares the wound bed, the AHSC is distributed and spread within the wound (Fig. 7).[79] Product application does not require surgical application, which allows for outpatient therapy. The AHSC forms points of skin that expand and initiate wound closure from multiple areas of epithelialization instead of just from the wound margins. In addition, the pluripotent nature of the hair follicle stem cell allows the cells to become skin appendages upon new growth, anecdotally creating a skin graft that can feel and sweat, which would obviously be a huge advantage.

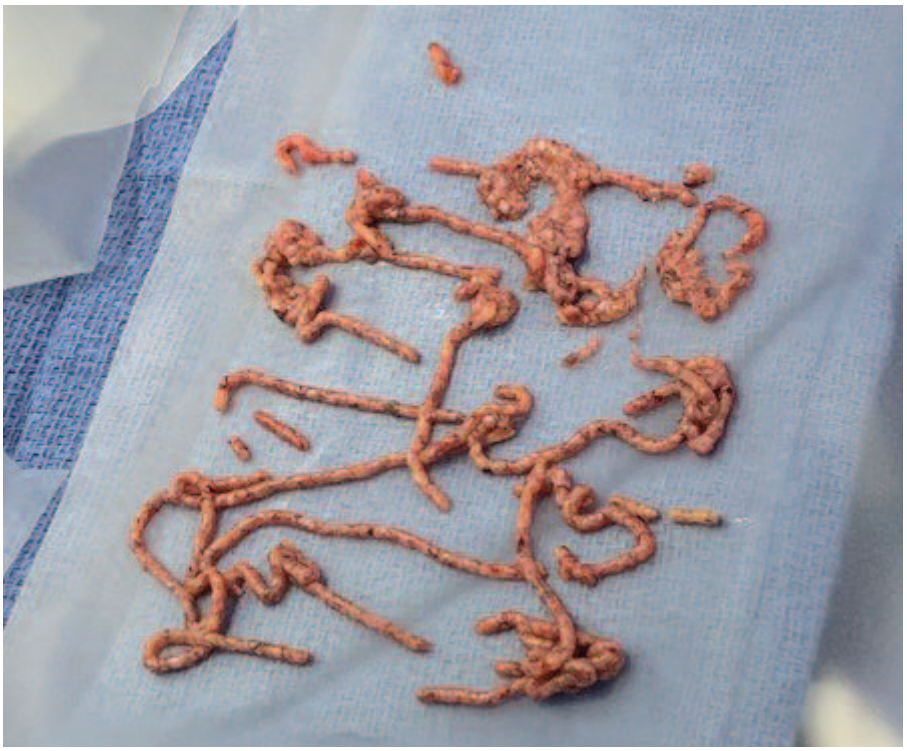

Figure 6. AHSC product prior to application (Photo provided by PolarityTE, Inc., Salt Lake City, UT, USA).

Armstrong et al. conducted a single-arm open-label feasibility study from November 2018 to May 2019 to treat 11 adult patients with Wagner 1 or 2 diabetic foot ulcers (DFU). The primary efficacy endpoint was rate of closure at 12 weeks following AHSC treatment. For application, wounds were cleaned and debrided followed by AHSC application. Application sites were then covered in multi-layer dressing and compression wrap. All 11 DFUs had graft take at 1 week after a single application of AHSC. Ten of the 11 (91%) completely closed within 8 weeks of application. The average wound closure rate for all 11 patients was 30 days and only 1 application of AHSC was required.[79]

As noted, this method is being further studied in a randomized control trial of AHSC versus standard of care. In the interim analysis, 50 total patients with Wagner 1 DFU were divided into an AHSC + standard of care group and a standard of care-only group (control). Standard of care involved offloading of DFU, debridement and wound care covering and/or compression wraps. The AHSC group involved full-thickness harvest and AHSC preparation and application as detailed above. There were

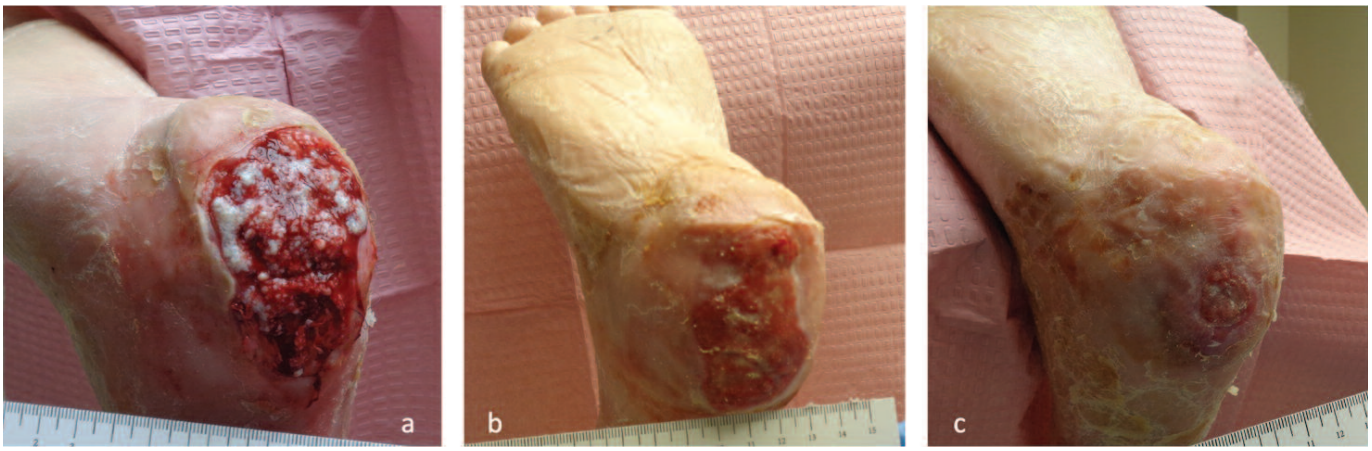

Figure 7. 22 cm² diabetic heel ulcer; a. AHSC deployment; b. 7 days post-AHSC; c. Closed 60 days post-AHSC. (Photo provided by PolarityTE, Inc., Salt Lake City, UT, USA).

significantly more wounds closed in the AHSC group compared to the control group (72% vs. 32%). There were no serious product-related adverse effects in the AHSC group. There was no significant difference in adverse events between the AHSC and control groups. Overall, AHSC seems to be promising in the interim analysis and may become a viable treatment option for patients in the future.[80]

Additionally, numerous case reports have demonstrated the utility of AHSC for more than just diabetic and venous leg ulcers. A 16-year-old patient with a large dehisced and irradiated wound bed after resection of a lower-extremity synovial sarcoma with exposed tendon was closed at 8 months post-operatively with complete functionality using AHSC, which demonstrated its potential to close complex wounds.[81] A 10-year-old boy who had been previously treated for flame burns with STSG had extensive contracture and keloid scars two years later. Scar tissue from the affected portion of his left chest was excised and treated with AHSC. There was notably less wound bed contraction and he had regenerated full-thickness and pigmented skin in the wound area, highlighting a reconstructive application of AHSC.[82] Two additional pediatric cases—a lower-extremity degloving injury and a surgical site wound, unfortunate sequelae after infection—were treated with AHSC and showed the successful regeneration of functional full-thickness skin complete with hair follicles and sweat glands.[83] This ability of cells to reorient in the correct layers and regenerate appendages such as hair follicles, glands and nerves, gives hope that this therapy may deliver truly functional dermis and epidermis. As in skin grafting, wound bed preparation will remain a key to making this product effective.

3D PRINTING

More recently, reports from Wake Forest have described novel work on the treatment of extensive wounds using a "mobile skin bioprinting system" with the help of image guidance.[12] That group used cellular therapy as an alternative to non-cellular biologic products. Using bioprinting, Albanna et al. was able to deliver either allogenic or autologous fibroblasts and keratinocytes to target sites in full-thickness excisional wounds. In their study, inkjet printing was used with imaging guidance to create a mobile skin bioprinting system. Using wound topography, the printer delivered cells into patient wounds in situ.[12] Both wound contraction and re-epithelialization were measured. From weeks 5 to 8, wounds treated with autologous cells (bioprinting) had significantly less contraction than untreated, matrix-treated and allogenic cell-treated wounds. By week 6, autologous-treated wounds had a significantly greater percentage of re-epithelization than untreated and matrix-treated wounds.[12] Overall, the work done by Albanna et al. has demonstrated the viability of bioprinting autologous cells onto patient wounds, which may one day be similar to the gold standard of STSGs.

Hakimi et al. demonstrated a handheld skin printer that can deliver consistent skin cell-laden sheets with equal thickness and composition in vivo. This printer would allow for the in situ formation of skin tissue sheets and would bypass other steps that are typically associated with regular bioprinters like washing, incubation and scanning of wound surfaces. Its ease of handling gives it a very relevant clinic application. This handheld printer's special bio-ink solution containing hyaluronic acid, fibrinogen, and type-I collagen has a suitable viscosity for printing and helps form important cross-linking between cell layers. Better mimicking of the spatial organization of intact tissues with the printer's biopolymers and cells helps improve the functionality of the tissue laid down. They reported successful bioprinting in murine and porcine excisional wound models.[84]

CONCLUSION

Skin expansion treatments are largely benefitting from the current medical advances in both skin composite cultures and pioneering technology, which continue to improve wound bed conditions and methods for laying down new epithelium. The criteria for a promising tissue-expansion treatment consistently seem to be that the product contains autologous cells with minimal donor-site morbidity from biopsy, is readily available or quickly processed, is easily applied (ideally avoiding the need for hospitalization or operating theaters in the case of chronic wounds) and produces epithelium most like native skin. For these reasons, the future looks promising for autologous homologous skin construct (AHSC), autologous skin cell suspension (ASCS) and 3D printing. These methods show immense potential for tackling two issues regarding denuded skin: the availability of a decent product and ease of application.

Sheets or slurries of CEAs do not have the same structural integrity as true skin. 3D printing can address some of the structural concerns that are currently being addressed by engineered composites of cellular or acellular, biologic or synthetic skin substitutes. The printers that use fibroblasts and keratinocytes along with crosslinking biomaterials lay down consistent sheets that better mimic true skin and can be tailored to specific wounds. With their cost and availability as major factors, a product that could be expeditiously applied in a clinical setting for chronic wounds and at bedside for burns, which doesn't require a large donor harvest or long incubation time, has great appeal.[84] Like 3D printing, ASCS shows real promise; it allows for wide and complete application, with an ability to reach all deep structures. On the other hand, ASCS has the limitation of only providing an epidermal layer; therefore, it is best used in conjunction with a different dermal regenerative strategy.

AHSC is a product based upon stem cells from the hair follicle. These pluripotent stem cells have the theoretical capacity to regenerate skin appendages. Patients who were treated with this product prior to its withdrawal from the market have been noted to gain skin that sweats and has light touch sensation. The questions of where and when to best use these products as part of a comprehensive plan of care are still unanswered. Better engineering does not have to come from a 3D printer alone. Research on modified skin substitutes has come a long way and further investigation into composite cultures, such as optimizing the polymer combinations in scaffolds, improving cross-linking for added strength, or the addition of mesenchymal stem cells, smooth muscle or endothelial cells, could improve a products likeness to real skin.[85] For 3D printing or composite cultures, the use of allografts or at least donor keratinocytes would expedite care for faster wound coverage. In order to ensure it provides a permanent graft, research into genetically modified cells to overcome immunologic rejection holds promise as well. STI

AUTHORS' DISCLOSURES

The authors declare that there are no conflicts of interest.

REFERENCES

1. Sen CK, Gordillo GM, Roy S, et al. Human skin wounds: a major and snowballing threat to public health and the economy. Wound Repair Regen 2009 Nov-Dec;17(6):763-71.
2. Han G, Ceilley R. Chronic Wound Healing: A Review of Current Management and Treatments. Adv Ther 2017 Mar;34(3):599-610.
3. Greenhalgh DG. Management of Burns. N Engl J Med 2019 Jun 13;380(24):2349-59.
4. American Burn Association. Burn Incidence Fact Sheet. .
5. Jeschke MG, van Baar ME, Choudhry MA, Chung KK, Gibran NS, Logsetty S. Burn injury. Nat Rev Dis Primers 2020 Feb 13;6(1):11.
6. Hop MJ, Polinder S, van der Vlies CH, Middelkoop E, van Baar ME. Costs of burn care: a systematic review. Wound Repair Regen 2014 Jul-Aug;22(4): 436-50.
7. Leary T, Jones PL, Appleby M, Blight A, Parkinson K, Stanley M. Epidermal keratinocyte self-renewal is dependent upon dermal integrity. J Invest Dermatol 1992 Oct;99(4):422-30.
8. Bhardwaj N, Chouhan D, Mandal BB. Tissue Engineered Skin and Wound Healing: Current Strategies and Future Directions. Curr Pharm Des 2017;23(24): 3455-82.
9. Papini R. Management of burn injuries of various depths. BMJ 2004 Jul 17;329(7458):158-60.
10. Rowan MP, Cancio LC, Elster EA, et al. Burn wound healing and treatment: review and advancements. Crit Care 2015 Jun 12;19:243.
11. Sierra-Sánchez Á, Kim KH, Blasco-Morente G, Arias-Santiago S. Cellular human tissue-engineered skin substitutes investigated for deep and difficult to heal injuries. NPJ Regen Med 2021 Jun 17;6(1):35.
12. Albanna M, Binder KW, Murphy SV, et al. In Situ Bioprinting of Autologous Skin Cells Accelerates Wound Healing of Extensive Excisional Full-Thickness Wounds. Sci Rep 2019 Feb 12;9(1):1856.
13. Jones I, Currie L, Martin R. A guide to biological skin substitutes. Br J Plast Surg 2002 Apr;55(3):185-93.
14. Samsell B, McLean J, Cazzell S, Dorsch K, Moyer PM, Moore M. Health economics for treatment of diabetic foot ulcers: a cost-effectiveness analysis of eight skin substitutes. J Wound Care 2019 Sep 1;28(Sup9): S14-S26.
15. Puthumana PP. Through the mists of time: Sushrutha, an enigma revisited. Indian J Plast Surg 2009 Jul;42(2):219-23.
16. Hauben DJ, Baruchin A, Mahler A. On the histroy of the free skin graft. Ann Plast Surg 1982 Sep;9(3): 242-5.
17. Reverdin J-L. De la greffe epidermique. 1872: P. Asselin, successeur de Béchet jeune et Labé.
18. Ehrenfried A. Reverdin and Other Methods of Skin-Grafting. Boston Med Surg J 1909; 161:911-17.
19. Blair VP, Brown JB. The Use and Uses of Large Split Skin Grafts of Intermediate Thickness. Surg Gynecol Obstet 1929;49:82-97.
20. Goldwyn RM. Kazanjian & Converse's Surgical Treatment of Facial Injuries. Arch Surg 1975;110(2): 227.
21. Ameer F, Singh AK, Kumar S. Evolution of instruments for harvest of the skin grafts. Indian J Plast Surg 2013 Jan;46(1):28-35.
22. Padgett EC. Skin grafting and the "three-quarter"-thickness skin graft for prevention and correction of cicatricial formation. Ann Surg 1941 Jun;113(6):1034-49.
23. Clodius L. The classic reprint. Die Transplantation Betreffend by Prof. Otto Lanz. Plast Reconstr Surg. 1972 Oct;50(4):395-7.
24. Tanner JC Jr, Vandeput J, Olley JF. The Mesh Skin Graft. Plast Reconstr Surg 1964 Sep;34:287-92.
25. Meek CP. Extensive severe burn treated with enzymatic debridement and microdermagrafting: case report. Am Surg 1963 Jan;29:61-4.
26. Schlottmann F, Bucan V, Vogt PM, Krezdorn N. A Short History of Skin Grafting in Burns: From the Gold Standard of Autologous Skin Grafting to the Possibilities of Allogeneic Skin Grafting with Immunomodulatory Approaches. Medicina (Kaunas) 2021 Mar 2;57(3):225.
27. Cohen J. Earliest attempt at free skin grafting. Ann Plast Surg 1995 May;34(5):552-3.
28. Fowler G. On the transplantation of large strips of skin for covering extensive granulating surfaces, with report of a case in which human and frogskin were simultaneously used for this purpose. Ann Surg 1889;9: 179-91.
29. Fiakos G, Kuang Z, Lo E. Improved skin regeneration with acellular fish skin grafts. Engineered Regeneration 2020;1:95-101.
30. Yamamoto T, Iwase H, King TW, Hara H, Cooper DKC. Skin xenotransplantation: Historical review and clinical potential. Burns 2018 Nov;44(7):1738-49.
31. Sharma K, Ralston DR, Giblin V, MacNeil S. Engineering of Accepted Skin-Equivalent Tissue for Tissue Repair: Current State and Perspectives. Reference Module in Biomedical Sciences. 2019
32. Zhou J, He W, Luo G, Wu J. Fundamental immunology of skin transplantation and key strategies for tolerance induction. Arch Immunol Ther Exp (Warsz) 2013 Oct;61(5):397-405.
33. Hoekstra MJ, Kreis RW, du Pont JS. History of the Euro Skin Bank: the innovation of preservation technologies. Burns 1994;20 Suppl 1:S43-7.
34. Richters CD, Hoekstra MJ, du Pont JS, Kreis RW, Kamperdijk EW. Immunology of skin transplantation. Clin Dermatol 2005 Jul-Aug;23(4):338-42.
35. Burd A, Chiu T. Allogenic skin in the treatment of burns. Clin Dermatol 2005 Jul-Aug;23(4):376-87.
36. Dai C, Shih S, Khachemoune A. Skin substitutes for acute and chronic wound healing: an updated review. J Dermatolog Treat 2020 Sep;31(6):639-48.
37. Phillips TJ, Gilchrest BA. Clinical applications of cultured epithelium. Epithelial Cell Biol 1992 Jan;1(1):39-46.
38. Beele H, Naeyaert JM, Goeteyn M, De Mil M, Kint A. Repeated cultured epidermal allografts in the treatment of chronic leg ulcers of various origins. Dermatologica 1991;183(1):31-5.
39. Fratianne R, Papay F, Housini I, Lang C, Schafer IA. Keratinocyte allografts accelerate healing of split-thickness donor sites: applications for improved treatment of burns. J Burn Care Rehabil 1993 Mar-Apr; 14(2 Pt 1):148-54.
40. Rheinwald JG, Green H. Serial cultivation of strains of human epidermal keratinocytes: the formation of keratinizing colonies from single cells. Cell 1975 Nov;6(3):331-43.
41. Grafting of burns with cultured epithelium prepared from autologous epidermal cells. Lancet 1981 Jan 10;1(8211):75-8.
42. Atiyeh BS, Costagliola M. Cultured epithelial autograft (CEA) in burn treatment: three decades later. Burns 2007 Jun;33(4):405-13.
43. Ronfard V, Rives JM, Neveux Y, Carsin H, Barrandon Y. Long-term regeneration of human epidermis on third degree burns transplanted with autologous cultured epithelium grown on a fibrin matrix. Transplantation 2000 Dec 15;70(11):1588-98.
44. Gallico GG 3rd, O'Connor NE, Compton CC, Kehinde O, Green H. Permanent coverage of large burn wounds with autologous cultured human epithelium. N Engl J Med 1984 Aug 16;311(7):448-51.
45. Green H. The birth of therapy with cultured cells. Bioessays 2008 Sep;30(9):897-903.
46. Dean SJ, Nieber S, Hickerson WL. The use of cultured epithelial autograft in a patient with idiopathic pyoderma gangrenosum. Ann Plast Surg 1991 Feb; 26(2):194-5.
47. Gallico GG 3rd, O'Connor NE, Compton CC, Remensnyder JP, Kehinde O, Green H. Cultured epithelial autografts for giant congenital nevi. Plast Reconstr Surg 1989 Jul;84(1):1-9.
48. Leigh IM, Purkis PE. Culture grafted leg ulcers. Clin Exp Dermatol 1986 Nov;11(6):650-2.
49. Eldad A, Burt A, Clarke JA, Gusterson B. Cultured epithelium as a skin substitute. Burns Incl Therm Inj 1987 Jun;13(3):173-80.
50. Ronfard V, Rives JM, Neveux Y, Carsin H, Barrandon Y. Long-term regeneration of human epidermis on third degree burns transplanted with autologous cultured epithelium grown on a fibrin matrix. Transplantation. 2000 Dec 15;70(11):1588-98.
51. Guilbaud J. Problems Created by the Use of Cultured Epithelia. In: Masellis M, Gunn SWA, eds. The Management of Burns and Fire Disasters: Perspectives 2000. Dordrecht: Springer. 1995.
52. Chester DL, Balderson DS, Papini RP. A review of keratinocyte delivery to the wound bed. J Burn Care Rehabil 2004 May-Jun;25(3):266-75.
53. Vericel Corporation. Epicel: Cultured Epidermal Autografts. www.epicel.com/what-is-epicel.html. Accessed 8/5/2022.
54. Hickerson WL, Remmers AE, Recker DP. Twenty-Five Years' Experience and Beyond with Cultured Epidermal Autografts for Coverage of Large Burn Wounds in Adult and Pediatric Patients, 1989-2015. J Burn Care Res 2019 Feb 20;40(2):157-165.
55. Fearmonti RM. Efficacy of Epidermal Skin Grafts Over Complex, Chronic Wounds in Patients With Multiple Comorbidities. Wounds 2016 Jul;28(7):226-32.
56. Hulsey A, Linneman P, Litt J. Clinical Usage and Economic Effectiveness of a Recently Developed Epidermal Autograft Harvesting System in 13 Chronic Wound Patients in a University-Based Wound Center. Cureus 2016 Nov 14;8(11):e878.
57. Miyanaga T, Kishibe M, Yamashita M, Kaneko T, Kinoshita F, Shimada K. Minced Skin Grafting for Promoting Wound Healing and Improving Donor-Site Appearance after Split-Thickness Skin Grafting: A Prospective Half-Side Comparative Trial. Plast Reconstr Surg 2019 Aug;144(2):475-83.
58. Hunyadi J, Farkas B, Bertényi C, Oláh J, Dobozy A. Keratinocyte grafting: a new means of transplantation for full-thickness wounds. J Dermatol Surg Oncol 1988 Jan;14(1):75-8.
59. Migliano E, Bellei B, Govoni FA, Bucher S, Picardo M. Fat and epidermal cell suspension grafting: a new advanced one-step skin regeneration surgical technique. J Exp Clin Cancer Res 2014 Feb 24;33(1):23.
60. Fraulin FO, Bahoric A, Harrop AR, Hiruki T, Clarke HM. Autotransplantation of epithelial cells in the pig via an aerosol vehicle. J Burn Care Rehabil. 1998 Jul-Aug;19(4):337-45.
61. Navarro FA, Stoner ML, Park CS, Huertas JC, Lee HB, Wood FM, Orgill DP. Sprayed keratinocyte suspensions accelerate epidermal coverage in a porcine microwound model. J Burn Care Rehabil 2000 Nov-Dec;21(6):513-8.
62. Wood FM, Kolybaba ML, Allen P. The use of cultured epithelial autograft in the treatment of major burn wounds: eleven years of clinical experience. Burns 2006 Aug;32(5):538-44.
63. Marston WA, Ennis WJ, Lantis JC 2nd, et al. Baseline factors affecting closure of venous leg ulcers. J Vasc Surg Venous Lymphat Disord 2017 Nov;5(6): 829-835.e1.
64. Kirsner RS, Marston WA, Snyder RJ, Lee TD, Cargill DI, Slade HB. Spray-applied cell therapy with human allogeneic fibroblasts and keratinocytes for the treatment of chronic venous leg ulcers: a phase 2, multicentre, double-blind, randomised, placebo-controlled trial. Lancet 2012 Sep 15;380(9846):977-85.
65. Lantis JC 2nd, Marston WA, Farber A, et al. The influence of patient and wound variables on healing of venous leg ulcers in a randomized controlled trial of growth-arrested allogeneic keratinocytes and fibroblasts. J Vasc Surg 2013 Aug;58(2):433-9.
66. Fiona Wood Foundation. https://www.fionawoodfoundation.com/fiona-wood. Accessed 8/05/2022.
67. Holmes IV JH, Molnar JA, Carter JE, et al. A Comparative Study of the ReCell® Device and Autologous Spit-Thickness Meshed Skin Graft in the Treatment of Acute Burn Injuries. J Burn Care Res 2018 Aug 17;39(5):694-702. doi: 10.1093/jbcr/iry029.
68. Iman A, Akbar MA, Mohsen KM, et al. Comparison of intradermal injection of autologous epidermal cell suspension vs. spraying of these cells on dermabraded surface of skin of patients with post-burn hypopigmentation. Indian J Dermatol 2013 May; 58(3):240.
69. Li Y, Zhang J, Yue J, Gou X, Wu X. Epidermal Stem Cells in Skin Wound Healing. Adv Wound Care

(New Rochelle) 2017 Sep 1;6(9):297-307.
70. Avita Medical, Inc. U.S. Food and Drug Administration Approves Expanded Use of the RECELL® System for the Treatment of Extensive Burns and Pediatric Patients. https://recellsystem.com/about-recell-system/ Accessed 2021.
71. Gravante G, Di Fede MC, Araco A, et al. A randomized trial comparing ReCell system of epidermal cells delivery versus classic skin grafts for the treatment of deep partial thickness burns. Burns 2007 Dec;33(8):966-72.
72. Hayes PD, Harding KG, Johnson SM, et al. A pilot multi-centre prospective randomised controlled trial of RECELL for the treatment of venous leg ulcers. Int Wound J 2020 Jun;17(3):742-752.
73. O'Neill TB, Rawlins J, Rea S, Wood F. Treatment of a large congenital melanocytic nevus with dermabrasion and autologous cell suspension (ReCELL®): a case report. J Plast Reconstr Aesthet Surg . 2011 Dec;64(12):1672-6.
74. Zweifel CJ, Contaldo C, Köhler C, Jandali A, Künzi W, Giovanoli P. Initial experiences using noncultured autologous keratinocyte suspension for burn wound closure. J Plast Reconstr Aesthet Surg 2008 Nov;61(11):e1-4.
75. RenovaCare. Our Technology: SkinGun and CellMist technology overview. https://renovacareinc.com/technology/.2022.
76. Esteban-Vives R, Choi MS, Young MT, et al. Second-degree burns with six etiologies treated with autologous noncultured cell-spray grafting. Burns 2016 Nov;42(7):e99-e106.
77. Esteban-Vives R, Corcos A, Choi MS, et al. Cell-spray auto-grafting technology for deep partial-thickness burns: Problems and solutions during clinical implementation. Burns 2018 May;44(3):549-59.
78. Svensjö T, Yao F, Pomahac B, Eriksson E. Autologous keratinocyte suspensions accelerate epidermal wound healing in pigs. J Surg Res 2001 Aug;99(2):211-21.
79. Armstrong DG, Orgill DP, Galiano R, et al. Complete wound closure following a single topical application of a novel autologous homologous skin construct: first evaluation in an open-label, single-arm feasibility study in diabetic foot ulcers. Int Wound J 2020 Oct;17(5):1366-1375.
80. Armstrong DG, Orgill DP, Galiano R, et al. A multicentre, randomised controlled clinical trial evaluating the effects of a novel autologous, heterogeneous skin construct in the treatment of Wagner one diabetic foot ulcers: Interim analysis. Int Wound J 2022 Jan;19(1):64-75.
81. Bade Y, Duarte-Bateman D, Manrique M, et al. SkinTE for the Treatment of a Complicated Wound after Synovial Sarcoma Resection: A Case Report. Plast Reconstr Surg Glob Open 2021 Aug 23;9(8): e3764.
82. Mundinger GS, Patterson CW; Co-Author: Abstract: Replacement of Contracted Split-Thickness Skin Graft and Keloid Scar with a Self-Propagating Autologous Skin Construct (SkinTE™). Plast Reconstr Surg Glob Open 2018 Sep 26;6(9 Suppl):95-95.
83. Isbester K, Wee C, Boas S, Sopko N, Kumar A. Regeneration of Functional, Full-Thickness Skin With Minimal Donor Site Contribution Using Autologous Homologous Skin Construct. Plastic Surgery Case Studies. 2020;6.
84. Hakimi N, Cheng R, Leng L, et al. Handheld skin printer: in situ formation of planar biomaterials and tissues. Lab Chip 2018 May 15;18(10):1440-51.
85. Nicholas MN, Jeschke MG, Amini-Nik S. Methodologies in creating skin substitutes. Cell Mol Life Sci 2016 Sep;73(18):3453-72.

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