SECTION VIII: USE OF SKIN SUBSTITUTES

Major advances in care have resulted in a marked decrease in mortality and also morbidity, especially with massive burns. In addition to survival the current focus in burn care is on improving the long term function and appearance of the healed or replaced skin cover as well as quality of life.

This focus on quality has generated a significant interest in the use of skin substitutes to be used to improve wound healing, to control pain, to more rapidly close a burn wound, to improve functional and cosmetic outcome, and, in the case of massive burns, to increase survival.

To more effectively address these new roles, the new generation of skin substitutes is biologically active. The bioactivity can modulate the burn wound instead of just covering the wound. The new products to be discussed have not displaced the more inert standard burn wound dressings but rather are used in conjunction and for quite specific indications.

The skin substitutes are initially classified according to whether they are to be used as a **temporary wound covering** to decrease pain and augment healing or a **permanent skin substitute** to add to or replace the remaining skin components.

The ideal properties and indications for these products will be better clarified after a discussion of the function of normal skin and the effect of a burn on skin integrity.[1-18]

A. ROLE OF BIOACTIVE SKIN SUBSTITUTES

The major stimulus for advances in skin substitutes is to improve the quality of the closed burn wound, control pain and avoid poor quality skin.[18-20]

Temporary skin substitutes can improve the healing while decreasing pain in superficial burns once the blisters and non-viable tissue has been removed. [16,17] In deeper burns the dead tissue will cause an inflammatory response producing both local and systemic effects. The systemic effects include a profound increase in metabolic rate with a marked increase in muscle wasting, and impairment in immune defenses.[1,2] Controlling this systemic response, by earlier removal of the dead burn tissue and closure of the wound, has markedly decreased overall mortality morbidity.[1l-13]

Permanent skin substitutes can then be used to permanently close the excised burn, especially in large burns where there isn't enough remaining skin to completely close the wound.[18-21]

The addition of **biological activity** to the skin substitute, improves the healing process with the intention of more rapidly healing a superficial burn and restoring valuable dermal components in a deeper burn wound bed thereby minimizing scarring and optimizing function.[18-21] A list of the non-cellular components of dermis, used in available skin substitute, is shown below.[22-26] Human epidermal and dermal cells are usually also added to dermal elements in permanent skin substitutes in addition to these dermal components.

DERMAL COMPONENTS STIMULATING HEALING[22-26]
• Structural component or scaffolding • Biologically active component stimulating all phases of healing • Collagen (protein) - Scaffold for cell migration and matrix deposition - Cell guidance • Elastin (protein) - Tissue elasticity • Fibronectin (protein) - Cell to cell adherence - Contact orientation for cells - Increases epithelial cell division, migration - Chemo attractant for fibroblasts, macrophages • Growth Factors (proteins) Stimulate all phases of wound healing • Glycosaminoglycan (glycosylated protein) - Cell adherence properties - Conduit for healing factors - Deactivator of proteases - Scaffold or foundation for dermal elements • Hyaluronic Acid (complex carbohydrate) - Maintaining matrix hydrated - Decreases inflammation - Stimulates healing - Proper cell alignment

B. AVAILABLE BIOACTIVE SKIN SUBSTITUTE

A list of skin substitutes, categorized by biologic make-up, is presented below. All have some degree of biologic activity for improving the wound-healing environment. **A disadvantage of all of these skin substitutes is the absence of active antimicrobial activity. However, early effective wound closure does decrease the risk of infection.**

AVAILABLE BIOLOGICALLY ACTIVE SKIN SUBSTITUTES

- Naturally occurring tissues
  Cutaneous allografts
  Cutaneous xenografts
  Amniotic membranes
  Porcine small intestinal submucosa
- Composite Synthetic-Biological
- Collagen based dermal analogs
  Integra
- Culture-derived tissue
  Bilayer human tissue
  Cultured autologous keratinocytes
  Fibroblast seeded dermal analogs
  Epithelial seeded dermal analog

Skin substitutes can also be categorized as to use and indication into temporary or permanent.
Temporary skin substitutes are used to
- Help heal the partial thickness burn (or donor site)
- Close the clean excised wound until skin is available for grafting

There are typically no living cells present.
Permanent skin substitutes are used
- To replace lost skin providing either epidermis or dermis, or both
- To provide a higher quality of skin than a thin skin graft

Most permanent skin substitutes contain viable skin cells as well as components of the dermal matrix.

C. TEMPORARY BIOACTIVE SKIN SUBSTITUTES

The purpose of a temporary skin substitute is twofold. Temporary skin substitutes are typically a bilayer structure. There is an outer epidermal analog and a more biologically active inner dermal analog.[18-21] The first objective is to close the wound, thereby protecting the wound from environmental insults.[18,19,27-29] The second objective is to provide an optimal wound healing environment by adding dermal factors which activate and stimulate wound healing.[18-29] Biologically active dermal components naturally are typically provided to the inner layer, which is then applied to the remaining dermis in a partial thickness burn or to an excised wound. Below is a list of the commonly available dermal matrix elements present in these products, and their actions.

IDEAL PROPERTIES OF A TEMPORARY SKIN SUBSTITUTE

- Rapid and firm adherence properties for closure of the wound
- Relieves pain
- Easily applied and secured
- Does not incite inflammation
- Stimulates wound healing
- Barrier to micro-organisms
- Avoids wound desiccation
- Optimizes healing environment
- Does not cause hypertrophic tissue response
- Haemostatic
- Prevents evaporative water loss
- Flexible yet durable
- Easy to remove when
  - Wound has re-epithelialized
  - Wound ready for grafting
- Cannot transmit disease
- Inexpensive
- Long shelf life
- Does not require refrigeration

The currently available products are listed below:

Available Bioactive Temporary Skin Substitutes						
Product	**Company**	**Tissue of Origin**	**Layers**	**Category**	**Uses**	**How supplied**
Human allograft	Skin bank	Human cadaver	Epidermis and dermis	Split thickness skin	Temporary coverage of large excised burns	Frozen in rolls of varying size
Pig skin Xenograft	Brennan Medical St. Louis, Mo	Pig dermis	Dermis	Dermis	Temporary coverage of partial thickness and excised burns	Frozen or refrigerated in rolls
Human amnion	On site procurement	Placenta	Amniotic membrane	Epidermis Dermis	Same as above	Refrigerator
Oasis®	Healthpoint, LTD San Antonio, Tx	Xenograft	Extracellular wound matrix from small intestine submucosa	Bioactive Dermal like Matrix	Superficial burns Skin graft donor sites Chronic wounds	Room T° storage Multiple sizes 3x3.5cm 7x20cm
Biobrane®	Dow Hickam/Bertek Pharmaceuticals	Synthetic with added denatured bovine collagen	Bilayer product outer silicone Inner nylon mesh with added collagen	Synthetic epidermis and dermis	Superficial partial thickness burns, Temporary cover of excised burns	Room T° storage 15x20inch 10x15cm 5x15inch 5x5 inch
Transcyte®	Smith and Nephew Wound Management Largo, FL	Allogenic Dermis	Bilayer product Outer silicone Inner nylon seeded with neonatal fibroblasts	Bioactive Dermal Matrix Components on Synthetic dermis and epidermis	Superficial to mid-Partial thickness burns Temporary coverage of excised burns	Frozen in 5x7.5 inch sheets

1. Human Allograft (Cadaver skin)

Human allograft is generally used as a split-thickness graft after being procured from organ donors.[30-32] When used in a viable fresh or cryopreserved state, it vascularizes and remains the "gold standard" of temporary wound closures. It can be refrigerated for up to 7 days, but must be stored frozen for extended periods. It is also used in a non-viable state after preservation in glycerol or after lyophilization: however, most existing data describe best results when it is used in a viable state. The epidermal component provides a barrier until rejected by the host in 3-4 weeks. The dermis revascularizes and incorporates.

Homograft, another term for human allograft, can only be obtained from a tissue bank as strict protocols are required for harvesting and storage. Donors must be rigidly screened for potential viral and bacterial disease to avoid any transmission of disease. The product is in limited supply and very expensive.

The primary indication for use is to cover a large excised burn wound until an autogenous skin or a permanent skin substitute becomes available. Allograft is also used to cover a wide meshed skin graft, sealing the interstices during the healing process.

Allograft Skin

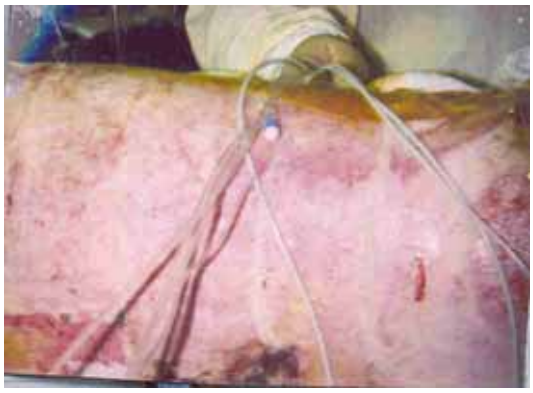
Advantages

- A bilayer skin providing epidermal and dermal properties
- Re-vascularizes maintaining viability for weeks
- Dermis incorporates into the wound

Disadvantages

- Epidermis will reject
- Difficult to obtain and store
- Risk of disease transfer
- Expensive
- Need to cryopreserve

Allograft Application to Excised Wound 8-1



Cadaver skin is nicely adherent to the wound

2. Xenografts

Although various animal skins have been used for many years to provide temporary coverage of wounds, only porcine xenograft is widely used today.[33-34] The epidermis of the porcine xenograft is removed and the split thickness dermis is provided in rolls. Split-thickness porcine dermis can be used after cryopreservation, or after glycerol preservation. It effectively provides temporary coverage of clean wounds such as superficial second degree burns and donor sites.[33,34] Porcine xenograft does not vascularize, but it will adhere to a clean superficial wound and can provide excellent pain control while the underlying wound heals.

In general, xenograft is not as effective as homograft but is more readily available and less expensive. Primary indications are for coverage of partial thickness burns during healing and used burn wounds prior to skin grafting.

Xenografts:

Advantages

- Good adherence
- Decreases pain
- More readily available compared to allograft
- Bioactive (collagen) inner surface with fresh product
- Less expensive than allograft

Disadvantages:

- Does not revascularize and will slough
- Short term use
- Need to keep the fresh product frozen

Current Use of Pigskin 8-2

Pigskin consists of a thin dermal layer (epidermis removed) which is stored frozen to maintain adhesive properties. The dermis is meshed to allow drainage to seep through

8-3

The pigskin dermis adheres to a cleaned partial thickness burn
- a dry gauze dressing follows

3. Human Amnion

Human amniotic membrane is used in many parts of the world as a temporary dressing for clean superficial wounds such as partial-thickness burns, donor sites, and freshly excised burns.[35,36] Amniotic membrane is generally procured fresh and used after brief refrigerated storage. It can also be used in a nonviable state after preservation with glycerol. Amnion does not vascularize but still can provide effective temporary wound closure. The principal concern with amnion is the difficulty in screening the material for viral diseases. The risks of disease transmission must be balanced against the clinical need and the known characteristics of the donor. The primary indications are the superficial burn and the excised wound.

8-4

Human Amnionic Membrane

Advantages

- Acts like biologic barrier of skin
- Decreases pain
- Easy to apply, remove
- Transparent

Disadvantages

- Difficult to obtain, prepare and store
- Need to change every 2 days
- Disintegrates easily
- Risk of disease transfer

4. Oasis Wound Matrix®

This product is made of the submucosa of the porcine small intestine found between the mucosa and muscularis, in the wall of the porcine small intestine.[37,38] The freeze dried cellular natural matrix retains its natural collagen and matrix structure and contains most of the bioactive matrix proteins found in the human dermis.

The submucosal layer is approximately 0.2mm in thickness but is quite durable. The product is freeze-dried removing the cells. The product is sterile, porous, biocompatible and non-immunogenic. It has a long shelf life and can be stored at room temperature. The OASIS® is incorporated into the wound bed over approximately 7 days and needs to be re-applied if the wound has not yet healed. The outer barrier function is diminished with incorporation.

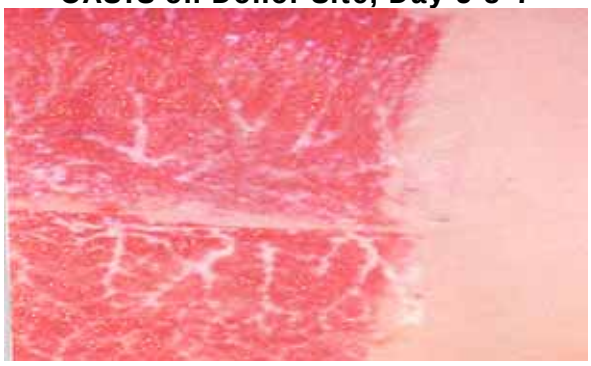
The primary indication is for use in difficult to heal non-burn wounds. Its use in burns is for the partial thickness burn and the skin graft donor site.

OASIS Application to Partial Thickness Burn 8-5

Upon application, product is moistened with saline, then covered by a non-adherent secondary dressing

OASIS Wound Matrix on Donor Site 8-6

OASIS on Donor Site, Day 5 8-7



Nearly totally re-epithelialized: matrix is incorporating with wound surface

8-8

Oasis Wound Matrix®

Advantages

- Excellent adherence
- Decreased pain
- Provides bioactive dermal like properties
- Long shelf life, store at room T°
- Relatively inexpensive

Disadvantages

- Mainly a dermal analog
- Incorporates and may need to be reapplied

5. Biobrane™

This product is a two-layer membrane.[39,40] The outer epidermal analog is constructed of a thin silicone film with barrier functions comparable to skin. Small pores present in silicone allow for exudates removal and has permeability to topical antibiotics.

The inner dermal analog is composed of a three-dimensional irregular nylon filament weave upon which is bonded type I collagen peptides. The surface binding of inner membrane is potentiated by collagen-fibrin bonds as well as fibrin deposition between the nylon weave. A thin water layer is maintained at the wound surface for epidermal cell migration maintaining moist wound healing.

Excellent adherence to the wound significantly decreases pain in the superficial partial thickness burns. The silicone and nylon weave provides flexibility. The biobrane is removed once the partial thickness wound has re-epithelialized or the covered excised burn wound is ready for grafting. However, if left in place for more than 2 weeks the product is difficult to remove as tissue grows into the inner layer. Biobrane L contains a nylon fabric woven from monofilament threads that provide a less dense matrix and less adherence, preferred e.g. on a donor site. There is likely very little direct bioactivity from the collagen peptides.[40] The product has a long shelf life and can be stored at room temperature. It is also relatively inexpensive.

The primary indication is for closure of the clean superficial burn or the excised burn wound.

8-9

Biobrane

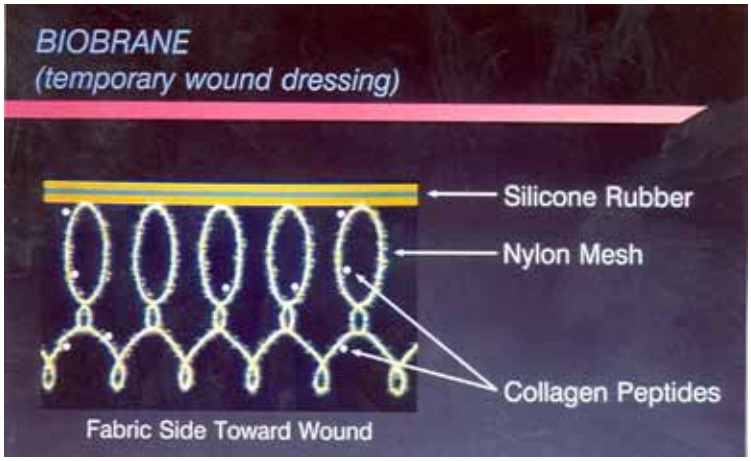
Advantages

- Bilayer analog
- Excellent adherence to a superficial burn
- Decreases pain
- Maintains flexibility
- Easy to store with long shelf life
- Relatively inexpensive

Disadvantages

- Has very little direct bioactivity
- Difficult to remove if left in place over 2 weeks

8-10



Components of Biobrane showing silicone layer and amino nylon mesh coated with collagen peptides

8-11

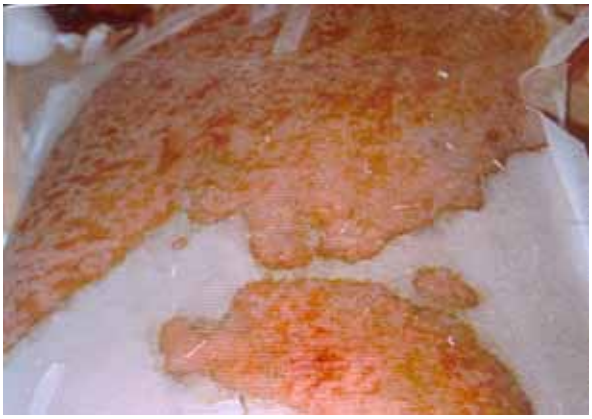
Outer surface shown. Note pores

8-12

Biobrane on modest stretch: surface smooth

Scald Burn 8-13

Biobrane on Debrided Scald 8-14



Day one: nicely adherent to wound

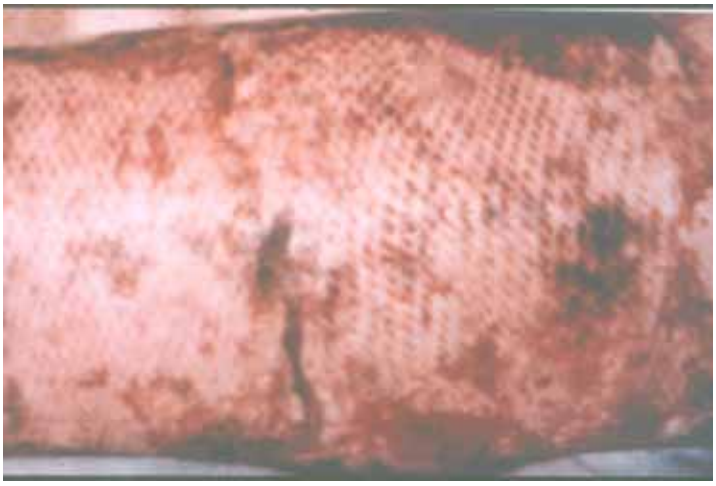
Superficial thigh burn 8-15

Covered with Biobrane (Day 10)

Biobrane removal 8-16

Opaque appearance indicates burn wound has re-epithelialized

Temporary Skin Substitute Covering Mesh Graft 8-17



The coverage avoids damage to the interstices as re-epithelialization occurs

6. TransCyte™

This product is also a bilayer skin substitute.[41,42] The outer epidermal analog is a thin nonporous silicone film with barrier functions comparable to skin. The inner dermal analog is layered with human neonatal foreskin fibroblasts which produce products mainly collagen type I, fibronectin and glycosaminoglycans.

A subsequent cryo-preservation destroys the fibroblasts but preserves the activity of fibroblast-derived products on the inner surface. These products do stimulate the wound healing process. A thin water layer is maintained at the wound surface for epidermal cell migration.

The nylon mesh provides flexibility and excellent adherence properties significantly decrease pain in the partial thickness burn. The product is peeled off after the wound has re-epithelialized. TransCyte must be stored at –70 C° in order to preserve the bioactivity of the dermal matrix products.

The primary indication is for closure of the clean superficial to mid-dermal burn, especially useful in children. TransCyte is also indicated for the temporary closure of the excised wound prior to grafting. Tissue growths tends to be less of a problem even if the product is kept in place for over two weeks.

8-18

TransCyte™

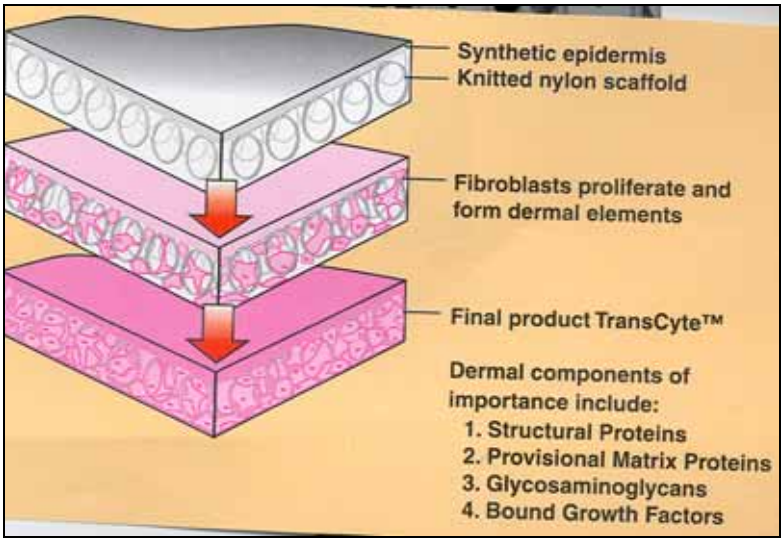
Advantages

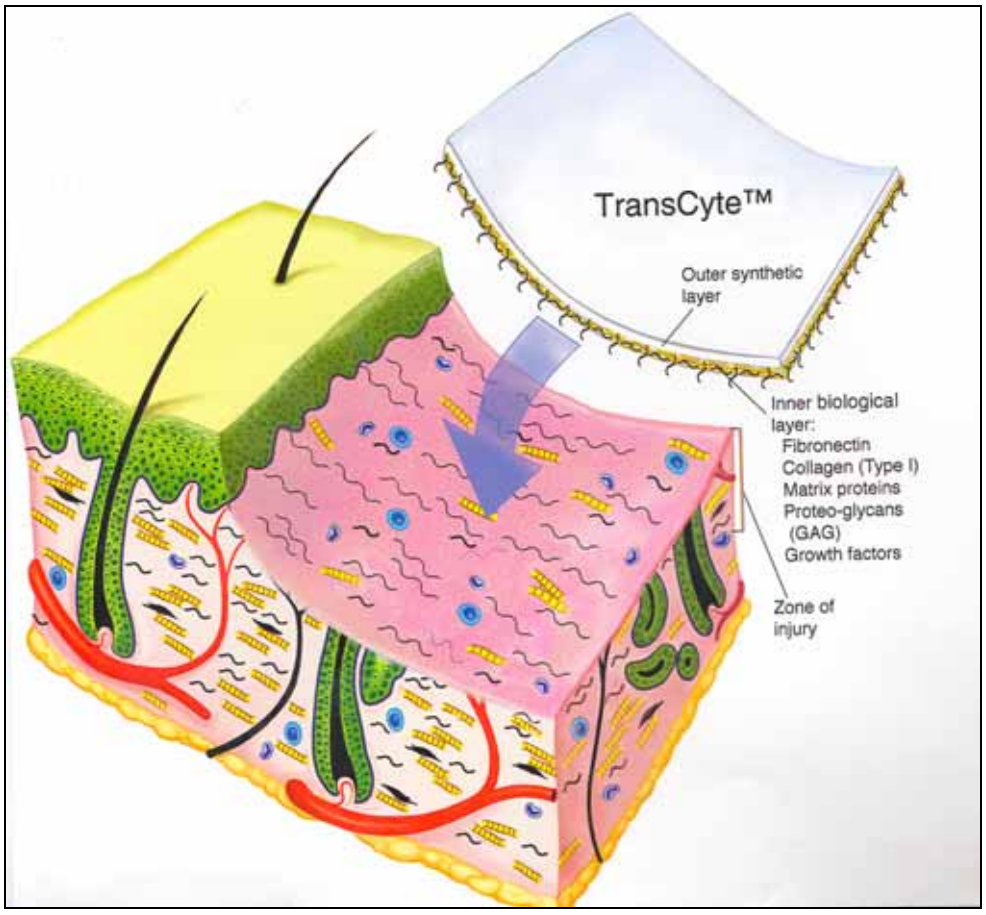
- Bilayer analog
- Excellent adherence to a superficial to mid-dermal burn
- Decreases pain
- Provides bioactive dermal components
- Maintains flexibility
- Good outer barrier function

Disadvantages

- Need to store frozen till use
- Relatively expensive

Components of TransCyte 8-19





Schema demonstrating the two-layer structure, the inner layer being bioactive.

TransCyte in Sealed Cassette 8-21

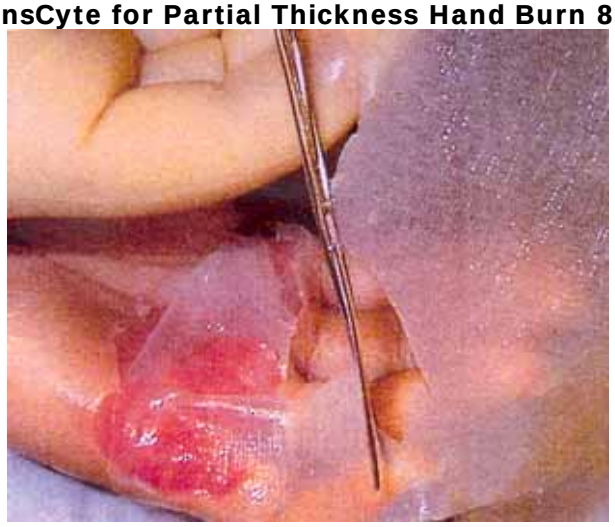
Stored at -70°Centrigrades

Debrided mid dermal burn.

Closed with skin substitute (Transcyte) cut to fit. Expect minimal to no scar using this approach.

8-22 8-23

TransCyte for Partial Thickness Hand Burn 8-24



Cutting the sheet to fit with a small overlap followed by initial immobilization until adherent

TransCyte on Foot Burn (3 days) 8-25

Note flexibility of the dressing

TransCyte on Leg Burn (10 days) 8-26

Opaque appearance indicating re-epithelialization beneath dressing for removal

TransCyte (Day 12) 8-27

Skin substitute being removed

D. PERMANENT SKIN SUBSTITUTES

The purpose of these products is to replace full thickness skin loss as well as to improve the quality of the skin, which has been replaced after a severe burn.[20-23]

As opposed to the bilayer concept of the ideal temporary skin substitute, permanent skin replacement is much more complex.

There are two approaches to developing a permanent skin substitute.[21-23] The first approach is the use of a bilayer skin substitute, with the inner layer being incorporated into the wound as a neodermis, rather than removed like a temporary product. The outer layer is either a synthetic to be replaced by autograft (epidermis) or actual human epithelial cells. The epithelial cells, which will form epidermis barrier function, is not often sufficiently developed at placement to act immediately as an epidermal barrier.

The second approach is the provision of either just an epidermal or a dermal analog, i.e. a one layer tissue. These products are technically not permanent skin substitutes upon initial placement as there is no bilayer structure.

Permanent Skin Replacement

- Bilayer structures with biologic dermal analog and either synthetic or biologic epidermal analog
- Skin components
  - Epidermal cells alone
  - Dermis alone
  - Co-culture of epidermal cells and fibroblasts

The ideal property as shown below would be that of a bilayer structure.

Ideal Properties of A Permanent Skin Substitute

- Rapid and excellent adherence properties
- Easily applied and secured to an excised wound
- Minimum wait period from time of burn to availability of skin substitute
- Bilayer tissue containing both epidermal and dermal eliminates to best replicate normal skin
- Rapid incorporation
- Cannot transmit disease
- Good functional and cosmetic result
- Inexpensive

The currently available clinical products are listed below. There are a number of permanent skin substitutes in the development stage, which will not be listed.

AVAILABLE PERMANENT SKIN SUBSTITUTES						
Product	**Company**	**Tissue of Origin**	**Layers**	**Category**	**Uses**	**How supplied**
Apligraf	Organogenesis , Inc and Novartis Pharmaceuticals Corp	Allogenic Composite	Collagen matrix seeded with human neonatal keratinocytes and fibroblasts	Composite: Epidermis and dermis	Chronic wounds, often used with thin STSG Excised deep burn	7.5cm diameter disc 1/pack
OrCel	Ortec International Inc.	Allogenic Composite	Collagen sponge seeded with human neonatal keratinocytes and fibroblasts	Composite: Epidermis and Dermis	Skin graft donor site, chronic wounds	6x6cm sheets
Epicel*	Genzyme Tissue Repair Corp	Autogenous keratinocytes	Cultured autologous keratinocytes	Epidermis Only	Deep partial and full thickness burns >30% TBSA	50cm^2 sheets in culture medium
Alloderm	Life Cell	Allogenic dermis	A cellular Dermis (processed allograft)	Dermis only	Deep partial and full thickness burns, Soft tissue replacement, Tissue patches	1x2cm to 4x12cm
Integra*	Integra Life Science Corp	Synthetic	Silicone outer layer on collagen GAG dermal matrix	BioSynthetic Dermis	Full thickness soft tissue defects definitive “closure” requires skin graft	2x2 inch 4x10 inch 8x10 inch 5/pack

1. Used mainly in burns

1. Epicel

This product, used mainly for very large burns is composed of the patients skin epithelial cells and referred to as cultured epithelial autograft (CEA). Therefore, only the epithelial layer is provided.[47-49] The product is made from a small biopsy of normal skin (2x2cm) from the burn patient. The epithelial cells are extracted and cultured. Use of a cell culture technique allows the keratinocytes to be grown in a thin sheet 10,000 times larger than the initial biopsy. This process does require 2-3 weeks from the time of biopsy. Often the burn wound is excised and covered with homograft (allograft) until the cells are ready to be transplanted. The CEA is then applied to the clean excised (or allograft covered) wound.

The CEA is supplied in sheets 2-6 cells thick on small pieces of petroleum gauze (50cm^2) which are bathed in culture medium. Immediate application is necessary. The CEA grafts are very fragile and easily rubbed off for at least several weeks. The backing is removed in several weeks as the CEA thickens and adheres. Graft-takes ranges from 30-75% of total epithelium applied. The epithelium gradually thickens but has a low resistance to sheer forces for some time. Application of allograft dermis, prior to CEA grafting appears to improve skin quality. In this case a dermal analog exists resulting in a bilayer skin. The primary indication is for very large burns.

Epicel

Advantages

- Patients own keratinocytes expanded several thousand fold
- Small skin biopsy required
- Can cover very large surfaces with reasonable graft take
- Used in large burns

Disadvantages

- 2 to 3 week lag time for production
- Provides only the epidermal layer
- Epithelial layer can be quite fragile for some time
- Needs to be used immediately on delivery
- Very expensive

Epicel Placement 8-28

The epidermal cells are placed on the wound on pieces of 50cm^2 petroleum gauze. The pieces are then secured to the wound bed and immobilized for 2-3 weeks

Epicel at 3 weeks 8-29

Note new epithelium (whitish patches) on the upper leg where the gauze has been removed

2. Alloderm

This product is basically treated human allograft with the epidermis removed.[50-52] The dermis is treated to produce a co preserved lyophilized allodermis, which incorporates. The product is used as a dermal implant. Therefore application of a thin epithelial autograft is required.

Primary indication is for use in the replacement of soft tissue defects. This product is not commonly used in large burns. A period of incorporation is required before the epithelial skin graft can be applied. The product has a long shelf life in its lyophilized form. It requires re-hydration prior to use.

Alloderm

Advantages

- Easy to store, an off the shelf product
- Does not require skin bank
- Comes in large and small pieces

Disadvantages

- Requires thin skin graft to provide epidermis
- Two procedures required to achieve bilayer skin
- Relatively expensive

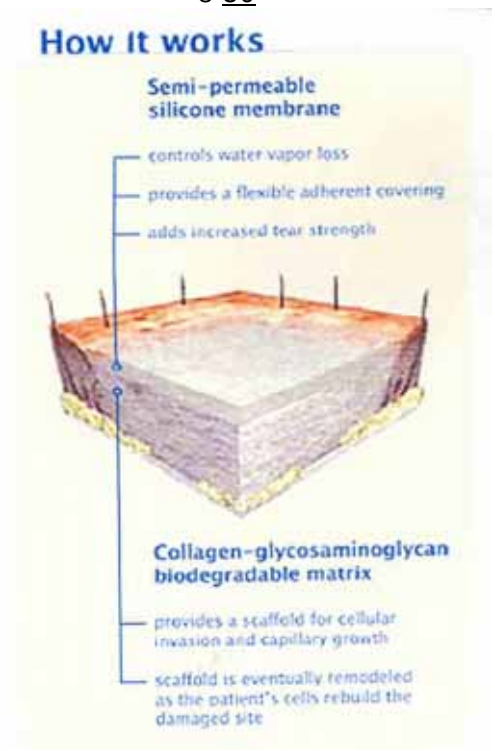
3. Integra

This product is composed of a dermal analog made of a biodegradable bovine collagen-glycosaminoglycan copolymer matrix The collagen and glycosaminoglycan is cross linked to attempt to maximize ingrowths of the patients own cells.[53-55]

The epidermal analog is a thin silicone elastomer providing temporary barrier protection. After the dermal analog incorporates and the surface revascularizes, at about 2-3 weeks, the silicone layer is removed and replaced with a very thin skin graft from the patient (or CEA cells). The Integra needs to be carefully immobilized for the first 2 weeks as movement will cause devascularization and loss of the product.

The primary indication is the treatment of large deep burns as well as reconstruction procedures. The incorporated neodermis appears to improve the function of the final skin once the epithelial graft is applied. The product is provided in a number of sizes and sheets stored in 70% isopropyl alcohol. Shelf life is very good.

8-30



The Integra Product 8-31

The two layer dermal analog is shown

Integra

Advantages

- Provides thick dermal analog
- Reasonable shelf life
- No risk of transmitting viruses
- Relatively inexpensive
- Used in large burns

Disadvantages

- Need to provide epidermis from the patient
- Dermal cells must come from the patient requiring product incorporation
- Two procedures required to achieve bilayer skin

SUMMARY

The scientific principles and practical approaches, to replacing skin either temporarily or permanently are advancing at a rapid rate. Much of these advances can be attributed to both advances in the field of bioengineering as well as increasing interest in optimizing the outcome of the burned skin.

The ideal properties of a bioactive temporary and a permanent skin substitute have been well defined. As expected, the properties of temporary skin substitutes are more concrete, easier to categorize and determine efficacy. A bilayer structures is the current standard with the dermal component being bioactive. Permanent skin replacement on the other hand is much more complex. A variety of approaches are being used which can be loosely categorized as either use of bilayer products (usually the outer layer to be replaced by epidermal autograft) or replacement of either dermal or epidermal elements separately. The terminology of the latter approach is difficult because these component products are really not permanent skin substitutes on initial application but become so only when all the elements are in place.

An understanding of the properties of each product is essential for the user to optimize outcome.

REFERENCES

1. Mast B: The Skin. In: Wound Healing. Cohen K, Diegelmann I, editors, WB Saunders. Philadelphia 1992:344-355.
2. Wright N, Allison M. The biology of epithelial cell populations. Clarendon press 1984:283-345.
3. Stenn S., Malhotra R. Epithelialization. In Wound Healing. Biochemical and Clinical Aspects. C Cohen (ed) WB Saunders, Philadelphia PA 1992:115-127.
4. Grillo H. Origin of fibroblasts in wound healing. Annals of Surgery 1963:157:453-467.
5. Karasck M. Mechanism of angiogenesis in normal and diseased skin.
6. Raghow R. The role of extracellular matrix in post-inflammatory wound healing and fibrosis. FASEB J 1994: 8:823-850.
7. Demling R. Burn care; in ACS Surgery. Wilmore D, ed NY, NY. Web MD 2002, p.479.
8. Herndon D. Total Burn Care. Saunders, Philadelphia 2002.
9. Neely A, Brown R, Chendening C, et al. Proteolytic activity in human burn wounds. Wound Rep Regen 1997:5; 302-9.
10. Young P, Grinnel F. Metalloproteinase Activation Cascade after burn injury: longitudinal analysis of the human wound environment. Journal of Investigative Dermatology 1994:103; 660-664.
11. Komgova R. Burn wound coverage and burn wound closure. Acta Chir Plast 2000:42; 64-68.
12. Sheriden R, Management of burn wounds with prompt excision and immediate closure. J Intensive Care Med 1994:9; 6-17.
13. Demling R, DeSanti L. Scar management strategies in wound care. Rehab Manage 2001:14; 26-32.
14. Still J. Primary excision of the burn wound. Clin Plast Surg 2000:27; 23-47.
15. Ladin D, Garner W, Smith D. Excessive scarring as a consequence of healing. Wound Repair Reg 1994:3; 6-14.
16. Scott P, Ghabary A, Chambers M et al. Biologic basis of hypertrophic scarring. Adv Struct Biol 1994:3; 157-165.
17. Erlich HP, Krummell T. Regulation of wound healing from a connective tissue perspective. Wound Repair Reg 1995:4; 203-210.
18. Badylak S. The extracellular matrix as a scaffold for tissue reconstruction. Cell Develop Biol 2002:13; 377-383.
19. Jones L, Currie L, Martin R. A guide to biological skin substitutes. Br J Plast Surg 2002:55(3); 185-193.
20. Gallico GG. Biologic skin substitutes. Clin Plast Surg 1990: 512-26.
21. Sheridan R, Tompkins R. Alternative wound coverings in: Total Burn Care, ed Herndon D., publisher Saunders, Philadelphia 2003, p 712.
22. Clark R, Folkvard J, Wortz R. Fibronectins, as well as other extracellular matrix proteins mediate human keratinocytes adherence. Journal of Invest Dermatol 1985:84; 378-383.
23. Clore J, Cohan I, Diegelmann R. Quantitation of collagen types I and III during wound healing. Proceed Soc Exper Biol Med. 1979:161; 337-340.
24. Doillon C, Dunn M, Bender E, et al: Collagen fiber formation in repair tissue: Development of strength and toughness. Collagen and related research 1985: 481-485.
25. Takashima A, Grinnell F. Human keratinocytes adhesion and phagocytosis. Prompted by fibronectin. J Invest Derm 1984:83; 352-358.
26. Miller E, Gay S, Collagen structures and function in wound healing: biochemical and clinical aspects. Cohen K, Editor. Saunders, Philadelphia 1992: p 130.
27. Nowicki CR, Sprenger C. Temporary skin substitutes for burn patients: a nursing perspective. J Burn Care Rehab 1988:9(2); 209-215.
28. Shakespeare P. Survey: use of skin substitute materials in UK burn treatment centers. Burns 2002:28(4); 295-7.
29. Smith K, Rennie MJ. Management of burn injuries: a rationale for the use of temporary synthetic substitutes? Prof Nurse 1991: 571-4.
30. Bondoc CC, Burke JF. Clinical experience with viable frozen human skin and a frozen skin bank. Ann Surg 1971:174; 371-82.
31. Herndon DN. Perspectives in the use of allograft. J Burn Care Rehab 1997:18; 56.
32. May SR, Still JM Jr., Atkinson WB. Recent developments in skin banking and the clinical uses of cryopreserved skin. (Review) J Med Assoc GA 1957:73; 233-6.
33. Song IC, Bromberg BE, Mohn MP, Koehnlein E. Heterografts as biological dressings for large skin wounds. Surgery 1966:59; 576-83.

34. Elliott RA Jr, Hoehn JG. Use of commercial porcine skin for wound dressings. Plast Reconstr Surg 1973:52; 401-5.
35. Ramakrishnan KM, Jayaraman V. Management of partial thickness burn wounds by amniotic membrane: a cost-effective treatment in developing countries. Burns 1997:23 (Suppl 1); 533-6.
36. Ganatra MA, Durrani KM, Method of obtaining and preparation of fresh human amniotic membrane for clinical use. J Pakistan Med Assoc 1996:46; 126-8.
37. Brown-Estris M, Cutshall W, Hiles M. A new biomaterial derived from small intestinal submucosa and developed into a wound matrix device. Wounds 2002:14; 150-166.
38. Demling R, Niezgoda J, Haraway G, Mostow E. Small intestinal submucosa wound matrix and full thickness venous ulcers. Wounds 2004:16; 18-23.
39. Smith DJ Jr. Use of biobrane in wound management. J Burn Care Rehab 995:16; 317-20.
40. Yang J, Tsai Y. Clinical comparison of commercially available Biobrane preparations. Burns 1989:15; 197-203.
41. Purdue G, Hunt J, Still M, et al. A multicenter clinical trial of a biosynthetic skin replacement, Dermagraft-T compared with cryopreserved human cadaver skin for temporary coverage of excised burn wounds. J Burn Care Rehab 1997:18; 52-7.
42. Demling RH, DeSanti L. Management of partial thickness facial burns (comparison of topical antibiotics and bio-engineered skin substitutes). Burns 1999:25; 256=261.
43. Bell YM, Falabella AF, Eaglstein WH. Tissue engineered skin. Current status in wound healing. Am J Clin Dermatol 2001:2; 305-13.
44. Folanga V, Sabolinski M. A bilayered living skin construct (APLIGFAF) accelerates complete closure of hard-to-heal venous ulcers. Wound Repair Regen 2000:7; 201-7.
45. Fivenson DP, Scherschun L, Choucair M, Kukuruga D, Young J, Shwayder T. Graftskin therapy in epidermolysis bullosa. J Am Acad Dermatol 2003:48; 886-92.
46. Still J, Glat P, Silverstein P, Griswold J, Mozingo D. The use of a collagen sponge/living cell composite material to treat donor site burn patients. Burns 2003:29(8); 837-41.
47. Sheridan RL, Tompkins RG. Cultured autologous epithelium in patients with burns of ninety percent or more of the body surface. J Trauma 1995:38; 48-50.
48. Rue LW III, Cioffi WG, McManus WF, Pruitt BA Jr. Wound closure and outcome in extensively burned patients treated with cultured autologous keratinocytes. J Trauma 1993:34; 662-7.
49. Loss M, Wedler V, Kunzi W, Meuli-Simmen C, Meyer VE. Artificial skin, split-thickness autograft and cultured autologous keratinocytes combined to treat a severe burn injury of 983% of TBSA. Burns 2000:26; 644-52.
50. Buinewicz B, Rosen B. Acellular cadaveric dermis (AlloDerm): a new alternative for abdominal hernia repair. Ann Plast Surg 2004:52; 188-94.
51. Wax MK, Winslow CP, Andersen PE. Use of allogenic dermis for radial forearm free flap donor site coverage. J Otolaryngol 2002:31; 341-5.
52. Druecke D, Steinstraesser L, Homann HH, Steinau HU, Vogt PM. Current indications for glycerol-preserved allografts in the treatment of burn injuries. Burns 2002:28; Suppl 1; S26-30.
53. Wisser D, Rennekampff HO, Schaller HE. Skin assessment of burn wounds covered with a collagen based dermal substance in a 2-year follow-up. Burns 2004:30; 399-401.
54. Navsaria HA, Ojeh NO, Moiemen N, Griffiths MA, Frame JD. Re-epithelialization of a full-thickness burn from stem cells of hair follicles micrografted into a tissue-engineered dermal template (Integra). Plast Reconstr Surg 2004:113; 978-81.
55. Frame JD, Still J, Lakhel-LeCoadou A, Carstens MH, Lorenz C, Orlet H, Spence R, Berger AC, Dantzer E, Burd A. Use of dermal regeneration template in contracture release procedures: a multicenter evaluation. Plast Reconstr Surg 2004:113; 1330-8.