Observations on the Development of an Artificial Skin: Presidential Address, 1982 American Burn Association Meeting

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The problem of wound closure following injury has been with us since complex organisms evolved, and the survival pressure to repair has been answered by a straightforward response—the open space is closed by bringing the wound edges together by contracture and not by the slower method of repair by regeneration of the lost tissue. It is not surprising that in the evolution of higher animals this direct method of preserving life was chosen. However, the more complex process of regeneration which would have the advantage of preserving function as well as life has always been the goal of medicine, and although striven for in the past, has even now achieved only limited success.

Injuries such as extensive third-degree burns may be taken as a case in point. Life is preserved by surgical wound closure, but function, both in the mechanical and cosmetic sense, may be permanently and disastrously lost because the missing tissue is replaced by scar which does not function like the lost skin. The next major improvement in the treatment of injury must be a more satisfactory replacement of lost or damaged tissue.

In the last few decades there have been increasing efforts to develop methods for replacing lost anatomic integrity and restoring function. In general, these efforts may be divided into those attempting biologic solution, based on actual regeneration of the part or transplantation from another individual; and a mechanical solution through mechanical replacement of destroyed tissue with a nonbiologic material such as is carried out in joint replacement. Unfortunately, there are significant problems with each. Regeneration of the lost tissue is surely the optimal goal for maintaining integrity and restoring function, but the basic information required to achieve this end does not exist at this time, and transplantation is seriously complicated by immunologic rejection. Mechanical materials, at least in certain areas, appear to offer a much more achievable functional replacement, based on a wide variety of new substances which are biologically inert. Although initially successful mechanically, these nonbiologic replacement parts have two serious problems. First, they do not take part in the biologic system and therefore, in a sense, are excluded from it. For example, antibacterial defense within a prosthesis itself, or at the implant tissue interface, is seriously deficient; and clinical bacterial infection, although rare, can pose a serious problem. Second, over time, the material itself and its join to the patient change in physical character. There is, for instance, fatigue and fracture in a total hip replacement, or the loosening of fixation of an artificial hip in the pelvis or femur. In either case the joint fails in time. (In passing it is fair to say that the unprotected surface position of skin makes it especially vulnerable. The almost universal development of infection has proved a major reason for the failure of synthetic polymeric materials tried as skin substitutes.)

With these problems in mind, our attempts to develop a physiologically acceptable skin substitute were neither purely biological nor purely mechanical in approach. When we examined the problem in the light of the criteria which this artificial skin must meet (Table I), it seemed most reasonable to choose a solution that was mechanical in its initial interface with tissue, but was
TABLE 1

<table>
<thead>
<tr>
<th>Biologic Properties</th>
<th>Physical Properties</th>
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<tr>
<td>1) Absent inflammatory or foreign body reaction</td>
<td>1) Epidermal portion imperme able to bacteria</td>
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<tr>
<td>2) Facilitate migration of fibroblasts and microvascular cells</td>
<td>2) Control of pore structure in the dermal portion</td>
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<td>3) Synthesis of neo-dermal tissue</td>
<td>3) Modulate fluid flux</td>
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<td>4) Low or absent antigenicity</td>
<td>4) Preventing rigidity</td>
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<td>5) Controlled rate of biodegradation</td>
<td>5) Tear strength</td>
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<td>6) Nontoxic metabolites</td>
<td>6) Modulus of elasticity</td>
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<td>7) Reduce contraction</td>
<td>7) Surface energy</td>
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<tr>
<td>8) Heal without scar</td>
<td>8) Handling and suturing characteristics</td>
</tr>
<tr>
<td>9) Prevent infection</td>
<td>9) Peel strength</td>
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soon transformed into a biologic solution, through cell migration and connective tissue synthesis with biodegradation of the mechanical prosthesis itself—in short, a combination of the mechanical and biological concepts. The following requirements shaped this decision: 1) the need for a material that would remain functional throughout the entire life of the patient; and 2) the development of a material that would not only replace structure but also replace the function of the tissue lost, including the cosmetic function. To achieve these goals, we believed that even though the initial implanted material could be nonviable and have only mechanical similarities to the tissue to be replaced, it must evolve to a mature final stage where it would be able to take part in antibacterial action and interact in a nearly normal way with the surrounding tissues. Further, it must undergo the same constant biologic remodeling that all viable tissue undergoes throughout the animal’s life in order to avoid such problems as change in physical character with time, such as stiffness, cracking, or loosening.

In all of this work I take pleasure in noting the essential involvement of Ioannis Yannas, Professor of Mechanical Engineering at the Massachusetts Institute of Technology. Without his unique, fundamental, and essential contribution the development of a successful skin substitute would never have taken place.

In our initial attempts to outline a research procedure to achieve our goals, we made the following hypotheses.

1) The abnormalities of scarred skin reside totally in the abnormally reconstituted dermis, not in epidermal abnormalities. We therefore concentrated on methods aimed at dermal reconstruction, believing that the patient’s own epidermal cells would always be readily available to form an epidermis if a neodermis could be developed.

2) Native collagen polymerized into fibers as a single molecular species would not act as a satisfactory biomaterial because its chemical interaction with tissue components activated defense reactions and rapid biodegradation. However, a copolymer fiber of collagen and glycosaminoglycan would produce a tissue reaction which promoted cellular and vascular supporting structure development by way of cell migration and the synthesis of normal connective material.

3) The function of normal dermis is related to the three-dimensional array of collagen fibers, and loss of dermal function is related to a loss of these spatial arrangements rather than to abnormalities in collagen or glycosaminoglycan molecular structure. Collagen bundles arranged as in dermis provide normal dermal function while the same collagen bundles arranged as scar produce the abnormal function of scar. Therefore, the cell migration and connective tissue synthesis must create a structure similar to that of normal dermis. This, we believe, would occur if an artificial dermal template made of the collagen-glycosaminoglycan lattice were placed on a wound bed. The template would act as a scaffolding to guide the migrating cells from the wound bed and their newly synthesized connective tissue material in a desired three-dimensional pattern.

4) Finally, if the template could be made to resemble the normal three-dimensional architecture of dermis, the cellular elements and supporting microvasculature would be induced to populate the scaffolding and synthesize normal connective tissue. This would be laid down in a pattern directed by the artificially manufactured template and would resemble normal dermis. The artificial scaffolding itself would be slowly biodegraded as it was replaced by ‘self’-connective tissue. The final material would resemble dermis more than scar and function more like dermis than like scar.

Using these criteria, what has evolved? We have developed a bilayer membrane along the lines of normal skin in which the outer layer (initially silastic epidermis) provides a barrier function, excluding unwanted material such as bacteria, and retaining body fluids and colloids. The inner and very much thicker artificial dermal layer consists of a fibrillar lattice constructed of fibers made of a copolymer of helical bovine hide collagen and chondroitin 6-sulfate derived from shark cartilage. The pore structure, glycosaminoglycan content, and cross-linked density of this material are carefully controlled, providing the required tensile strength, elasticity, and tear quality, as well as the prescribed three-dimensional structure and rate of biodegradation. Further, the degradation products of this biomaterial are nontoxic. This bilayer provides the physical characteristics required for intimate adherence to the wound bed, resembling that found in freshly harvested split-thickness autograft. It has sufficient elasticity to move with the underlying wound bed, and the ability to retain sutures. It reacts positively with the cellular elements of the wound bed without inflammation, foreign body reaction, or immunologic rejection.

The silastic epidermal portion is temporary and is designed to be removed at a time of election based on individual patient needs. It is replaced by the patient’s own epidermal cells seeded when the silastic is removed.
and grown into a cornified epidermis in situ on the patient’s newly developing derived ‘neodermis.’ In a sense this uses the patient’s own growth factors and substrates as tissue culture media.

It seems reasonable to comment here that the ability to construct each layer of the bilayer membrane in a different fashion and of different material allowed an important simplification to be made which allowed achievement of the total design criteria of the artificial skin easier to accomplish. Each layer could be considered as a separate entity, and each could make separate contributions to the total number of characteristics required of the bilayer membrane. In our design, the major portion of the physical qualities, such as resistance to bacterial invasion, strength, and water transport, were assigned to the epidermal layer while the biologic characteristics required were contributed by the dermal portion.

In the development of the membrane each design characteristic needed required a separate series of experiments which were carried out in Professor Yannas’ laboratories at M.I.T. Examples are: the requirements for helical collagen, the exact glycosaminoglycan moiety to be used, the per cent of this glycosaminoglycan per unit of helical collagen, and the pore size (i.e., the size of holes in the material) in the dermal matrix itself. One developmental problem will be used as an example of the complexities of the system and the exactness with which the dermis must be constructed to function in a biologically acceptable way. Pore size, or the distance between one collagen-GAG bundle and the next in the open lattice structure of the dermal template, proved to be of considerable importance. When this problem was approached, the method for production of fibers of the composite material (collagen-GAG) which would not trigger the inflammatory reaction, as well as a method of constructing a material out of these collagen-GAG fibers, had been developed by Professor Yannas. Our initial attempts produced a material that was clearly compatible with tissue but did not support migration of fibroblasts, much less vascular structures. After a considerable amount of time and effort it became obvious that the problem rested not in the chemical structure of the material but in its physical distribution in space and that in order to promote migration of connective tissue and its vascular supporting structure from the surrounding normal tissue, the scaffolding into which the cells were to migrate must contain pores of a certain diameter; and further, that these pores must be continuous throughout the material and oriented in such a way that the resulting neodermis would contain a fiber orientation which resembled nor-
mal dermis. Figure 1 shows a histologic section of two examples of artificial dermis. The lower border of each is designed to interface with the wound bed itself. The upper portion interfaces with the silastic epidermis (not present). Both are constructed of the exact same fibrillar material, but the lower preparation (B) is more compacted, with a considerably smaller pore size, while the upper (A) is a more widely open lattice. The material (B) did not provide a satisfactory dermal implant because cellular invasion was irregular and slow, never reaching a uniform distribution of cells throughout the material, while A provided a scaffolding which supported rapid migration of cellular elements, vessels, and newly forming connective tissue elements in the vascular supporting structure throughout the dermal implant by about 2 weeks. Pore size and orientation of these pores, therefore, proved a very important variable although before our investigations pore size had not been recognized as a parameter of consequence.

Having developed a material, or neodermis, that acted as a satisfactory skin, we moved to human trials following approval by the Human Studies Committee of the Massachusetts General Hospital and the Massachusetts Institute of Technology, obtaining informed consent from each treated patient (or responsible family member) in the Burn Unit of the Massachusetts General Hospital or the Shriners Burns Institute in Boston. The artificial material was used clinically as a skin substitute for the first time on a patient in October of 1979.

We believe that demonstrating the effectiveness of an artificial skin is dependent on the satisfactory replacement of large areas of skin rather than on a several-square-centimeter test area, so we have attempted to limit our applications to patients with life-threatening injuries. For the most part we have tried to cover 15% or more of the body surface with the artificial skin. Moreover, we believe that in addition to providing satisfactory skin closure in the short term the artificial skin must also provide satisfactory skin function, including cosmetic function, for the remainder of the patient’s life. In our experience thus far, we have found the material to provide a satisfactory skin replacement whose cosmetic appearance gives promise for some control of scar formation. The long-term behavior of the artificial skin is as yet unknown. The longest followup shows the skin to be satisfactory, but it has only been in place for 2½ years.

In the experience gathered thus far, we have noted no evidence of an immunologic reaction, or rejection of the artificial material: neither a primary nor second set response has occurred. Inflammation is conspicuously absent and no foreign body reactions have developed. Several patients treated have had demonstrated burn wound
sepsis and bacteremia at the time of eschar excision and artificial skin grafting. These grafts have taken without the development of sepsis so that present evidence indicates that the artificial material supports defense in a manner similar to a viable split-thickness autograft. The complications seen in the material's use have been secondary to technical errors on application—hematoma formation under the artificial material, and the development of infection under the graft—and have been few in number. Technical problems have been both at the time of application and in the care of the graft (particularly the Silastic elements) in the postoperative period.

The artificial skin, as it is presently clinically used, is as seen in Figure 2, having roughly the same handling, wound-adherent, and draping qualities of split-thickness skin. It is grafted into the excised wound using the exact technique utilized for split-thickness grafts to achieve primary wound closure with carefully constructed sutures without gaps, and undergoes the same postoperative management and dressing techniques as sheet autograft. It has become clear that premature loss of the Silastic epidermis without prompt seeding with epidermal cells causes damage to the developing neodermis, granulation formation at its surface, and a decrease in the eventual functional and cosmetic result. Although it is possible to allow the Silastic dermis to remain in place about 50
days before removal, for practical purposes the sooner the Silastic is removed and epidermal cells placed after cellular ingrowth into the dermal template has taken place, the more likely an optimal functional and cosmetic result will occur.

The formation of a neodermis begins immediately after artificial skin grafting and is best illustrated by staged examples taken from biopsies of an adult whose third-degree burn covered 80% of the body surface. Figure 3 shows a histologic section of artificial skin and wound bed 3 days following grafting. The cellular and vascular migration is already taking place, with newly synthesized connective tissue element already present. The junction of wound bed and artificial graft is identified with arrows and the intimate relationship between the artificial dermis and the wound bed is seen. At 21 days post grafting, the artificial template has been completely populated with connective tissue elements and well-developed vessels. The Silastic epidermis has been removed in histologic processing and is not present in the figure.

Since the problem of developing a neodermis with characteristics resembling dermis rather than scar remains of paramount importance, it is worth exploring the extent to which this has been accomplished. In this regard, two histologic sections taken within a millimeter of each other provide insight into the ability of the neodermis to induce dermal-like structure rather than scar. Figure 3 shows a biopsy of artificial skin that was placed immediately following excision of a deep burn that extended well into the muscle. The base of the
excision was muscle, and the artificial skin was laid on the remaining muscle itself. The upper portion of the biopsy (A) has retained its neodermal quality. However, at the interface between the muscle and the neodermis there is a clearly developed scar (B), extending down into the muscle (C). This histologic difference between neodermal structure and scar is evident. It is also important to note that the scar deeply penetrates the muscle. This histologic appearance raises the possibility of conventional scar formation developing in the interface between normal tissue and the neodermis even though the dermal-like architecture is retained in the artificial material. Rotating and recutting the block deeper shows a different picture (Fig. 4). The neodermis (A) here sits comfortably on the muscle bed (B) without scar formation. We believe that the difference between the two sections was caused by incomplete debridement of thermally destroyed muscle in the original excision of burn eschar and scar formation in this small area of retained necrotic tissue following artificial skin grafting. A small distance away, however, the debridement was accurate, no necrotic tissue remained, and the interface between artificial dermis and deep tissue is without scar. This reinforces the importance of accurate debridement of all necrotic tissue as well as provides evidence for the absence of scar between artificial dermis and its normal graft bed.

Our experience to date has been developed in 36 burned patients, ranging in age from 2 to 90 years, who have
received artificial skin grafts in 68 separate surgical procedures, or an average of 1.8 artificial skin grafting operations per patient. The clinical behavior of artificial skin can be reasonably demonstrated by the following report. A 43-year-old man was admitted to the Massachusetts General Hospital 3 days following a 63% total body surface area burn, 50% of which was third degree. He was taken to the operating room on the first postadmission day, and the full-thickness eschar of the anterior trunk was excised and artificial skin placed on the chest and abdomen (Fig. 5). On the following day he underwent repeated eschar excisions and wound closure with autograft; the extent of artificial skin coverage was 20% of the body surface. Because of the lack of donor sites, the Silastic membrane was allowed to remain on the artificial skin for 30 days, was then removed and replaced with a thin-meshed epidermal graft. This took well and at 16 days post-epidermal grafting it formed a satisfactory epidermal closure. The functional quality of artificial skin at this time appeared excellent. Followup at 12 weeks after grafting shows continuing satisfactory closure, and its cosmetic and functional quality compares favorably with the area grafted at the same time with meshed autograft (Fig. 6).

In a second patient, who was admitted to the Shriners Burns Institute, Boston, the continuing quality of artificial skin at 6 months post grafting is shown in Figure 7. The area of artificial skin grafting is thicker, more pliable, and in the patient’s opinion more acceptable. In general, patients’ evaluation of the areas grafted with
Fig. 7. Area of artificial skin placed on the lower abdomen 6 months before picture surrounded by conventional meshed autograft. Improvement in texture and cosmetic appearance are evident.

artificial skin indicates that itching does not occur, their perception of sensation is at least as good and sometimes better than conventionally grafted areas, and they are happier with the cosmetic result.

Our experience is small and of short duration but I believe that our goal is providing an artificial skin physiologically acceptable to the human body and available 'off the shelf' for closing wounds of any size has been achieved. It is too soon to assess our success in reducing scarring and improving cosmetic and functional results, but we are tantalized and cheered by some of the early results.