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Basic healing phenomena around permanent percutaneous implants*

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Summary

Percutaneous implants represent foreign objects which penetrate a surgically created skin defect. The integument/implant interface generally fails to seal and heal and the device soon needs to be removed due to extrusion and infection.

During the past 10 years, experimental surgical and histological studies were undertaken at Clemson University to determine the etiology of implant failure. According to our findings and those of others, the following phenomena contribute to implant failure: extrusion, permigration, avulsion, infection and a combination of these.

Histologically, the implant/tissue interface healing phenomena were quantitatively the same in rabbits, dogs and goats, with only minor quantitative differences. Camouflaging the implant surface with denatured collagen did not promote fibrous-tissue formation but rather retarded its onset.

Histochemical evaluation of collagen and ground substance in the interstices of the porous implant revealed the following: collagen and ground substance differed both in quality and quantity from those found in the implant capsule or in adjacent normal subcutis. Both collagen and glycosaminoglycans undergo accelerated turnover and do not reach normal configurations and concentrations.

These findings may provide explanations to the described failure modes of marsupialization and permigration.

Introduction

Percutaneous devices are objects, usually of a non-biological material, which penetrate a surgically created integumental defect, thereby providing a connec-

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tion between the interior of the body and the external environment. They may serve a variety of purposes in clinical and experimental medicine. In maxillofacial reconstruction, they may serve as anchors for external facial contours or appendices to be fastened to internal structures such as the ears, nose and chin. They may also serve as conduits for signal or power conducting wires or for catheters.

There are only very few naturally occurring skin-penetrating objects and nature has not been successful in maintaining them in a permanent percutaneous location. The deer antler and mammals' teeth may serve as examples. The deer antler is a bony skeletal extension which breaks through the skin during the summer and is shed a few months later in an annual rhythm. Mammalian teeth have a lifelong pergingival survival if the periodontal tissues remain healthy, but they are eventually also extruded.

All classical prosthetic percutaneous devices have a common end point: they are extruded within days to weeks. Extreme surgical, medical and sanitary precautions in patients and experimental animals have prolonged the functional lifetime of these devices up to a year in the hands of a few investigators.

The clinical and experimental failures in maintaining long-term percutaneous connections have led a few research centers to study systematically the failure modes and to develop methods, materials and designs which would circumvent these failure modes [1]. Briefly, 5 failure modes have been identified which lead to extrusion of the device.

— Smooth surface implants are eventually encapsulated by proliferating epidermis which forms a sinus tract along the implant at a migration rate of 2 mm every week. This mode has been coined *marsupialization* [2].
— When the implant has a porous surface, allowing connective tissue and epidermis to migrate into the implant, the epidermis will migrate throughout the entire porosity and eventually encapsulate and surround it. This process is called *permigration* [3].
— Percutaneous devices are subjected to a variety of forces which can lead to mechanical disruption of the interface. This is known as *avulsion* [4].
— At any time during the implantation period, independent of, or supported by, the processes described above, contamination of the interface may, and usually will, lead to implant *infection* which cannot be resolved by medical or surgical treatment.
— Usually failure occurs due to a *combination of the above-mentioned failure modes*.

Histological studies by Winter led to his theory that epidermal migration (marsupialization) could be stopped by a barrier of normal collagen containing connective tissue [2]. Our subsequent studies at Clemson University indicated further that abnormal or immature collagen, as seen in a wound of less than 36 hours, will

not stop epidermal proliferation and migration. This may be the reason for sinus tract formation around percutaneous implants.

Because we believe that these qualitative differences of connective tissue might be the key to percutaneous failure, our laboratory undertook a series of histological studies of epidermal and subcutaneous healing around and into test implants of polyethylene terephthalate fabric in a velour configuration. These studies include the work of 5 graduate students who, over a period of the past 8 years, have contributed step-by-step to the development of our present understanding of percutaneous failures.

Materials and methods

Test implants made from commercially available, surgical quality polyethylene terephthalate velour were used in all experiments to allow for comparisons (Fig. 1). Velour sheets were cut into 2 × 1 cm strips. Two strips were glued together on their woven surfaces using medical-grade polyether polyurethane adhesive to create a double-surfaced velour test implant with an impermeable center. Test implants which were appropriately cleaned, sterilized in ethylene oxide and degassed for 8 days under a hood were implanted percutaneously on the backs of rabbits (Fig. 2), dogs and goats, such that approximately half of the length of the implant was placed subcutaneously and half of it protruded above the skin. Others were totally implanted in the subcutaneous tissue for comparison. After various time periods of up to 30 days, the implants with surrounding tissues were removed and processed for histological examination.

Some of the implants were treated before implantation with bovine tendon-derived denatured collagen (Fig. 3) and others with autologous serum [5]. The

Fig. 1: Surface scanning electron micrograph of the polyester fibers and interstices of polyethylene terephthalate velour. The fibers have an average diameter of 30 microns.

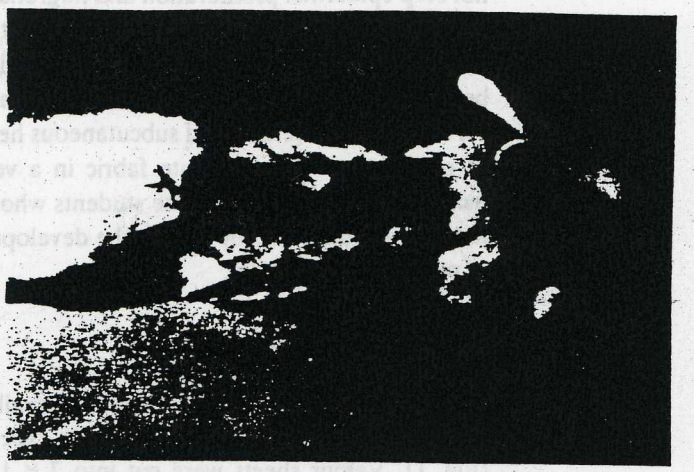

Fig. 2: A rabbit with 8 test and control implants in a percutaneous location on the back. The animal is wearing a protective elizabethan collar which helps prevent damage of the implant or implant site.

implant development, surgical and histological procedures have been described in detail elsewhere [5, 6].

Results

Species comparisons

Our comparative studies between the 3 animal species, i.e., goats, dogs and rabbits, revealed that all light-microscopically defined healing processes around and inside the velour were basically the same. However, there were differences in rates of epidermal migration, connective tissue formation and macrophage accumulation between the species. Goats showed more connective-tissue formation and maturation with fewer macrophages; rabbits had accumulated macrophages and showed less connective tissue formation and maturation at all studied time intervals; the dog results were always intermediate [6, 7].

These results indicate that there are probably no major qualitative differences of percutaneous healing between the studied mammals and that studies performed in these animals are equally acceptable for extrapolation to man. There may be differences in rates of healing, degree of connective-tissue maturation and occurrence of failure. Clinical observations of percutaneous implants appear to indicate that slower healing and prolonged functional implant life may be expected in man [1].

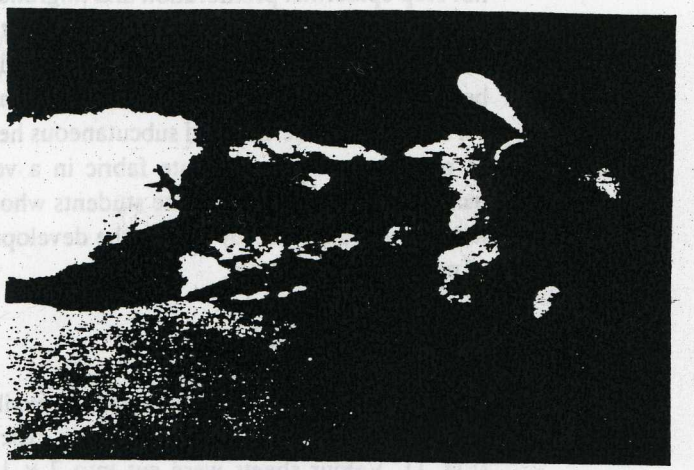

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Fig. 3: Polyethylene terephthalate velour impregnated with denatured bovine collagen slur; surface scanning electron micrograph; magnification × 128.

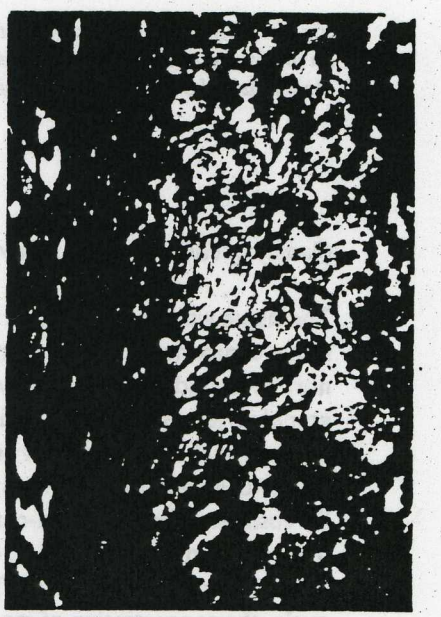

Fig. 4: Light micrograph (low-power magnification) of a hematoxylin-eosin stained tissue/implant interface of a dog. The connective tissue capsule is on the left; the polyethylene terephthalate velour implant extends from the middle of the picture (asterisk) to the right. The density of the connective tissue on the left reaches the implant but does not extend into it.

Fig. 5: Higher-power magnification of the same type of interface as shown in Figure 5 of an implant in a rabbit. The collagen organized in thick bundles in the capsule, to the left of the asterisk, shows very little organization in the velour interstices to the right of the asterisk.

Quality of connective tissue in velour

The tissue which migrates and proliferates into the velour initially consists of the same components and undergoes the same developmental phases found in normal wound healing. However, after the disappearance of polymorphonuclear granulocytes and the onset of fibroblastic proliferation, the collagen within the interstices of the implant is not produced in amounts and a quality usually expected and actually observed in the capsule which forms around the implant. Hematoxylin-eosin and Gomori's Tri-chrome stains indicate little collagen formation and differences in its quality (Figs. 4 and 5). The collagen fibers are thinner and never organized into thick bundles. This was observed at all time intervals. Simultaneously, the concentration of macrophages and giant cells gradually but steadily increased and actually appeared to displace connective tissue.

Epidermal basal cells were seen to migrate and proliferate into spaces formerly occupied by connective tissue components and displaced them with ease, eventually filling all spaces in the velour with the product of epidermal maturation, keratin [5, 6].

Fig. 6: Light micrograph of a polyethylene terephthalate velour implant in a rabbit 10 days after implantation. The tissue bed is to the right (W) outside of this picture. The polyethylene terephthalate fibers (triangles and arrows) are surrounded by the migrating epidermal cells. Because of the maturing process of epidermal cells, the fibers are eventually imbedded in cornified cell layers (triangles). The basal cells migrate into the space occupied by purely organized connective tissue (asterisk) in the left margin and left lower quarter of the picture. As can be seen in the middle of the picture (S), epidermis takes over the entire space available in the interstices between the fibers.

These results lead us to believe that the connective tissue which forms inside the spaces of the velour does not mature into strong scar tissue, as is observed in normal wound healing or in the capsule around the implant. Furthermore, we believe that macrophages and giant cells, stimulated by the presence of the foreign material, successfully compete for the available space around the polymer fibers, probably by devouring all connective tissue components. It is not clear whether macrophages are actually responsible for the lack of connective tissue maturation, whether they take advantage of this, or both. It has been demonstrated by others that stimulated macrophages can have both an inhibiting and a stimulating effect on fibroblast proliferation and activity [8, 9].

It became apparent that epidermal basal cell migration and proliferation is not prevented by this 'inferior' connective tissue or collagen (Fig. 6). Rather, the basal cells appear to be able to displace collagen and all other connective tissue components, perhaps by enzymatic activity. We have found that the rate of epidermal migration is inversely proportional to the maturity of the connective tissue. Consequently, epidermal marsupialization and permigration are allowed to proceed around and through percutaneous implants.

Collagen and autologous serum-impregnated implants

Stimulated by the work of others, we implanted collagen or serum-impregnated implants and studied them in the hope that these coatings would enhance fibroblast activity and promote connective tissue formation and maturation. Our findings indicated the contrary. The proliferation of connective tissue was actually retarded for up to 10 days by the presence of the denatured collagen and appeared to be dependent on prior lysis of the collagen coating. Serum treatment appeared to have no measurable effect when compared with untreated implants in the same animals [5].

Special studies on collagen and ground substance

When special stains and histochemical methods were applied in a presently ongoing study to analyze the extracellular structural components of the connective tissue in the velour, the following results were obtained. Collagen and reticulum remained scarce and disorganized throughout the observed implantation period (Figs. 7 and 8). While collagen production was diminished in the capsule

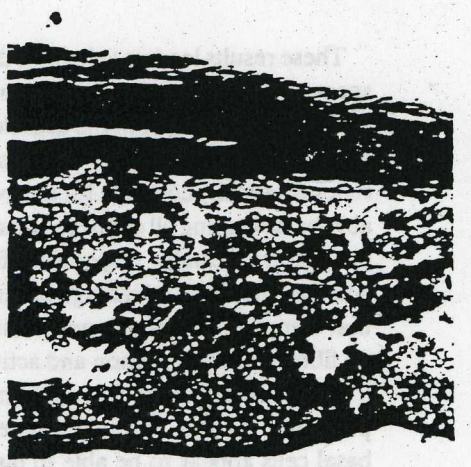

Fig. 7: Twenty-eight-day implant in a rabbit; Gomori's Tri-chrome stain. The stain indicates little collagen formation inside the interstices of the velour.

Fig. 8: Twenty-eight-day implant in a rabbit; Gomori's silver stain. The stain indicates minimal reticulum fiber development inside the interstices of the velour.

surrounding the implant by 28 days post-implantation, synthesis continued within the velour at a high rate (as demonstrated by leucine aminopeptidase sensitive stains). Since the collagen within the velour remained sparse, we interpret this as evidence of a continued high collagen turnover. The collagen fiber diameter, observed to be decreased in our specimens, has been linked by others to both the rate of collagen phagocytosis and the rate of mature collagen turnover [10].

It appears, however, that the formation and organization of collagen fibrils is, in part, also determined by the extracellular matrix. The extracellular matrix, or ground substance, contains, among other proteins, enzymes and cell metabolites, glycosaminoglycans (GAGs). This group of structural compounds, when abnormal, has been demonstrated to cause alterations in the normal aggregation patterns of precipitated collagen during in vitro testing [11, 12]. The presence of GAGs also appears to alter the kinetics of the aggregation process [13]. Furthermore, analyses of the composition of GAGs in prostheses capsules have shown both time dependency and variance from mature scars [14].

This evidence prompted our investigation into the composition of the non-collagenous extracellular matrix. The following findings were derived from test

implants which were totally implanted into the subcutaneous space in order to eliminate percutaneous variables. Significant differences were found in the composition and activity of the matrix within the implant interstices when compared with the capsule surrounding the implant, an incisional scar, and normal connective tissue from the same animal, as demonstrated with alcian blue/periodic acid schiff at a pH of 2.5 and β-glucuronidase. β-Glucuronidase has been linked to abnormal collagen formation in rheumatoid arthritis [15] and hydrolyses GAGs. β-Glucuronidase was found in the interstices of both the short (10-day) and long (28-day) implants. Used in this manner, the presence of the enzyme is indicative of the hydrolysis or turnover of GAGs.

In conclusion, it might be said that: connective tissue found within the interstices of polyethylene terephthalate velour implants differs both in quality and quantity from that found in the surrounding connective tissue; this collagen undergoes degradation or accelerated turnover; these changes occur simultaneously with alterations in the GAG fraction of the extracellular matrix. This information, when viewed in the light of the in vitro demonstration of the effects of GAGs upon the formation and organization of collagen fibers, suggests that the abnormal collagen found within the implants may be the result of continued phagocytosis and turnover of the collagen compounded by alterations in the formation of the collagen fibrils, and that these changes might be related to the presence of defective or altered ground substance.

Conclusion

Unfortunately and as usual, our findings raised more questions than they answered. Collagen and ground substance are both products of fibroblasts and do not reach a normal developmental stage and equilibrium between breakdown and build-up in this experimental situation. We know that macrophages share implant pores with fibroblasts, influence fibroblast activity (both through stimulation and inhibition) and devour the fibroblast products, collagen and ground substance. What still needs to be determined are the exact roles which the fibroblasts, macrophages and implant material play independently and in conjunction in the production of defective extracellular materials inside the implant pores.

Answers to these questions and progress in the synthesis of histocompatible polymers will eventually allow us to induce normal connective tissue scar formation at the implant skin interface. This, in turn, will result in a tenacious bond and an effective and lasting epidermal seal of the interface and permit us to permanently connect external devices to internal structures.

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