ACTA CHIRURGIAE PLASTICAE

INTERNATIONAL JOURNAL
OF PLASTIC SURGERY

VII · 3

1965

Acta chir. plast. 7:3:1965

CZECHOSLOVAKIA · PRAGUE · SZDN

Published four times (in 1959: two times) a year by Státní zdravotnické nakladatelství, (Czechoslovak Medical Press), Malostranské nám. 28, Praha 1. — Adress of the editorial office: Acta chirurgiae plasticae (M. Dobrkovský, M. D. — Secretary) Legerova 63, Praha 2, Czechoslovakia. — Orders through ARTIA, Smečky 30, Praha 2. — Press: Středočeské tiskárny, n. p., provoz 101, Hálkova 2, Praha 2. — A-22*51167

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The Hospital for Sick Children, Toronto (Canada)

THIN SKIN HEALING

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Wound healing has been studied and reviewed frequently (3, 6, 9, 11, 15, 19, 25, 34). Most research in this field has been carried out on animals. Those studies which have used human material have often relied on a few subjects or on a variety of wounds from varying locations. For these reasons and because of the importance of this subject to medicine and surgery, we have chosen to study skin-wound healing again.

Normal thin skin, as described by Ham and Leeson (17), is shown diagrammatically in Fig. 1. A break in the continuity of this structure, a skin wound, may result from a simple incision, an abrasion, a contusion, an avulsion or an excision of a piece of the skin. This study is confined to incised wounds.

A review of the literature reveals disagreement on the sequence of events which take place in normal skin healing. Gillman et al. (11) have presented a detailed description of the old and new concepts of skin incision healing (Fig. 2). The old concept is as follows:

1. Clot fills the space created by the incision and the fibrin strands of the clot hold the wound edges together.

2. Inflammatory cells, capillary buds and fibroblasts migrate from the wound walls and fill the wound space with granulation tissue.

3. New epidermis spreads from the periphery of the wound over this bed of fibrin clot and granulation tissue.

4. Collagen fibres are laid down in the granulation tissue after a lag period of a few days. The contraction of these fibres gradually squeezes capillaries and cells from the zone of repair which then becomes a white scar:

More recent investigators (9, 11) have critized the old concept of skin healing because it does not agree with their observations, and because it does not explain the facts that wounds heal better without clot and that punctate

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scars often form at suture holes. The new concept of skin healing is as follows (11):

1. The epidermal edges invariably fold inwards following incision.

2. There is usually little if any clot in the lower part of the wound after suturing. Clot delays healing when it is present.

Fig. 1. Diagram of normal skin. (A) Epidermis; (b) papillary layer of dermis; (c) reticular layer of dermis; (d) subcutaneous tissue; skin appendages; (e) sweat gland, and (f) hair follicle.

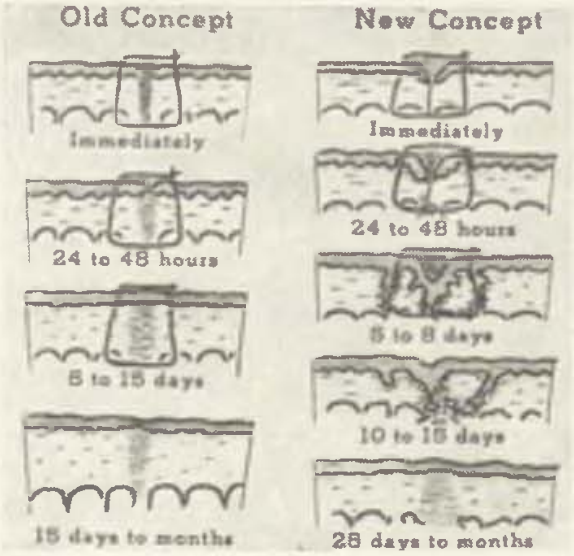

Fig. 2. Diagrammatic outline of the old and new concepts of skin healing adapted from Gillman et al.[11] Old Concept. Immediately, clot fills wound space; 24 to 48 hours, inflammatory elements and fibroblasts migrate into wound space, new epidermis proliferate over wound; 5 to 15 days, collagen fibres laid down; 15 days to months, collagen fibres mature and contract. New Concept. Immediately, epidermal edges invert, minimal clot in lower part of wound; 24 to 48 hours, epidermis migrates down wound walls to reach base as shown at 5 do 8 days, epidermis proliferates into defects of wound edges and suture tracts, fibroplasia begins in the subcutaneous and papillary layers; 10 to 15 days, epidermis retreats upwards as fibroplasia continues, often leaving islands; 25 days to months, epidermis becomes elevated and thin, collagen fibres thicken and contract, cells and vessels diminish in number.

3. Epidermal cells begin migrating down the walls of the wound under clot and debris during the first day of healing. The advancing sheets of thinned-out epidermis meet in the base of the wound at about 48 hours postoperatively. The epidermis, after re-establishing its continuity, invades defects in the walls of the wound and grows into suture tracts which are held gaping by the horizontal tension of the suture.

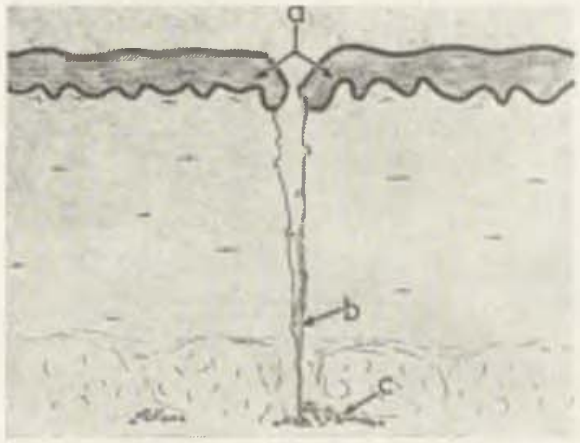

Fig. 3. Composite diagram of healing incisions during the first six postoperative hours. (a) Epidermal margins inverted; (b) early clot, and (c) subcutaneous hemorrhage.

Tab. 1. Description of Subjects Used in this Study and their Diagnoses

Subjects: 27 children with a total of 36 cutdown incisions		
Sex:	Male	14
	Female	13
Age:	1 day to 2 weeks	8
	2 weeks to 1 month	7
	1 month to 1 year	3
	1 year to 5 years	9
Principal diagnosis:		
	Congenital anomaly —	
	cardiac	9
	other	10
	Infection	6
	Malignancy	1
	Fibrocystic disease	1

4. Fibroplasia commences after epidermal continuity has been established. Active connective-tissue proliferation has begun in the subcutaneous tissue by about the fifth to eighth day of healing. The granulation tissue thus formed expands upwards into the wound. Connective tissue proliferates downwards from the papillary layer of the dermis at about two weeks postoperatively. The reticular layer of the dermis takes little part in fibroplasia.

5. As fibroplasia continues, the epidermis retreats upwards and often leaves islands of epidermal cells behind. Multinucleated giant cells and macrophages cluster around these islands and around the epidermal projections into the dermis. These epidermal projections and islands are gradually removed and their place is taken by new fibrous tissue.

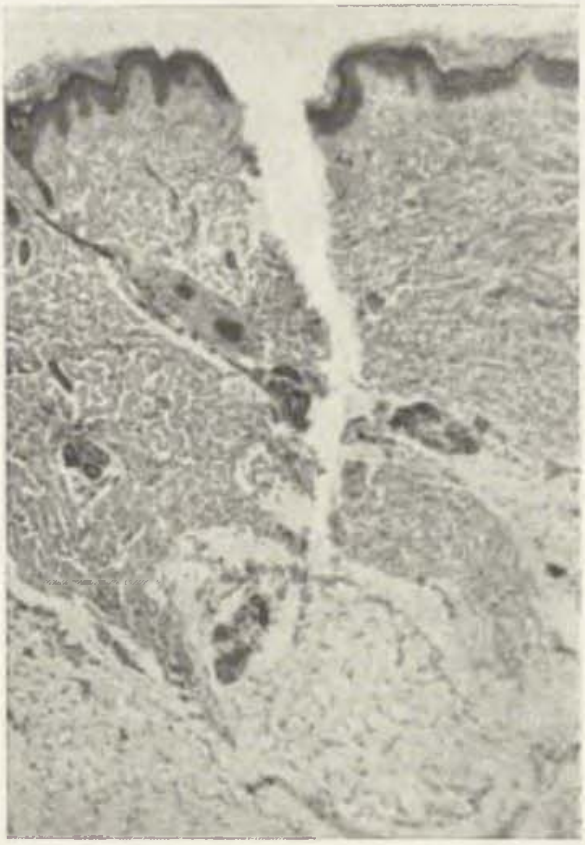

Fig. 4. Photomicrograph of skin incision five hours postoperatively. This section shows less subcutaneous hemorrhage than usual. Note. All sections illustrated here were cut from paraffin imbedded blocks and stained with hematoxylin and eosin.

6. More new collagen fibres are laid down old fibres become thickened. The repair area is very vascular and cellular at first, but as the scar matures and contraction takes place the vessels and cells become more sparse. Correspondingly, on gross examination, the pink raised scar becomes a flat white scar.

METHOD

With the above review as a background, a study of the healing of incised wounds in thin skin was carried out. The subjects used were children coming to autopsy with a transverse cutdown incision over the long saphenous vein at the ankle (Table I).

The cutdown incisions had been closed with plain catgut skin sutures. Sections were prepared by excising the region of the cutdown incision, fixing

it in 10% formol saline, dehydrating it in ascending strengths of ethyl alcohol, passing it through xylol, embedding and cutting it in paraffin, and staining it with hematoxylin and eosin. All sections were between 5 and 7 μ. in thickness. The zone of disruption caused by the polyethylene intravenous tube passing through the centre of the wound was avoided by cutting sections close to either end of the incision and at a distance from suture tracts.

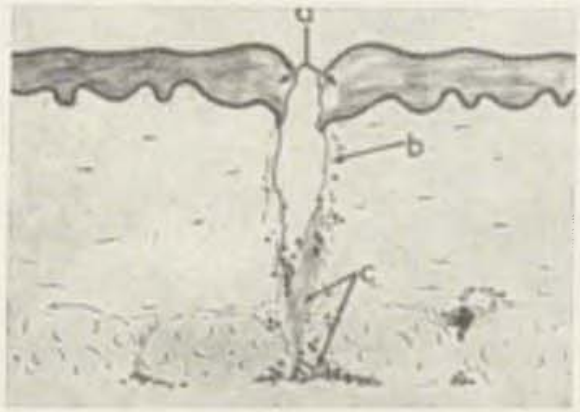

Fig. 5. Composite diagram of healing incisions from the period six hours to two days postoperatively. (a) Epidermal thickening; (b) homogenous coagulum lining wound walls, and (c) further inflammatory reaction.

The advantages of this experimental material were that it was easily obtained, it came from a constant site on the body, and it was taken from subjects with an age range from 1 day to 3½ years. The possible disadvantages of the experimental material were that it was potentially infected, it was obtained up to one day after death, and it was obtained from patients suffering from a variety of diseases and receiving a variety of medications.

However, infections if present were minimal and appeared to have little influence on the rate of healing. Tissues taken one day postmortem did not seem to be significantly altered because the subjects were refrigerated between death and autopsy. The variety of diseases and medications of subjects did not seem to cause variation in the rate of healing, but may have contributed to the uniformly slower rate of healing in our study as compared with that of Gillman et al. (**11**).

OBSERVATIONS AND DISCUSSION

A careful study of our experimental material has led us to the following scheme of thin skin healing.

Zero to Six-Hour Period

Fig. 3 illustrates the histological picture of the healing incision during the period immediately to six hours postoperatively. A representative section taken during this period is shown in Fig. 4. The epidermal margins are slightly inverted into the wound. This inversion is probably due to suturing and the

deforming force produced by cutting some of the supporting dermal collagen and elastic fibres while leaving others intact. A small amount of hemorrhage occurs at this stage from the divided subpapillary and subcutaneous vascular networks. This blood usually pools in the subcutaneous tissues between the loose connective tissue fibres where it may be a factor in initiating the inflammatory response. The early exudation of plasma and lymph rapidly jells into a fibrin network with red blood cells and leukocytes trapped in its interstices.

Six-Hour to Two-Day Period

This stage of healing (Fig. 5) is characterized by thickening of the epidermis along the margin of the incision, by the formation of a homogenous coagulum lining the wound walls, and by further fibrinous exudation and invasion by inflammatory cells. Fig. 6 shows a representative histological section at this stage. The clot was thought by early workers to function as a wound adhesive and a scaffold upon which epidermis migrated and into which capillaries and cellular elements grew. Recent workers believe that the clot is unnecessary and probably detrimental since it must eventually be phagocytosed or extruded (10). It is still believed to play a minor part in healing, as an early wound adhesive (25) and protective cover (11).

Fig. 6. Photomicrograph of skin incision two days postoperatively showing a fibrinous and cellular exudate in the wound space.

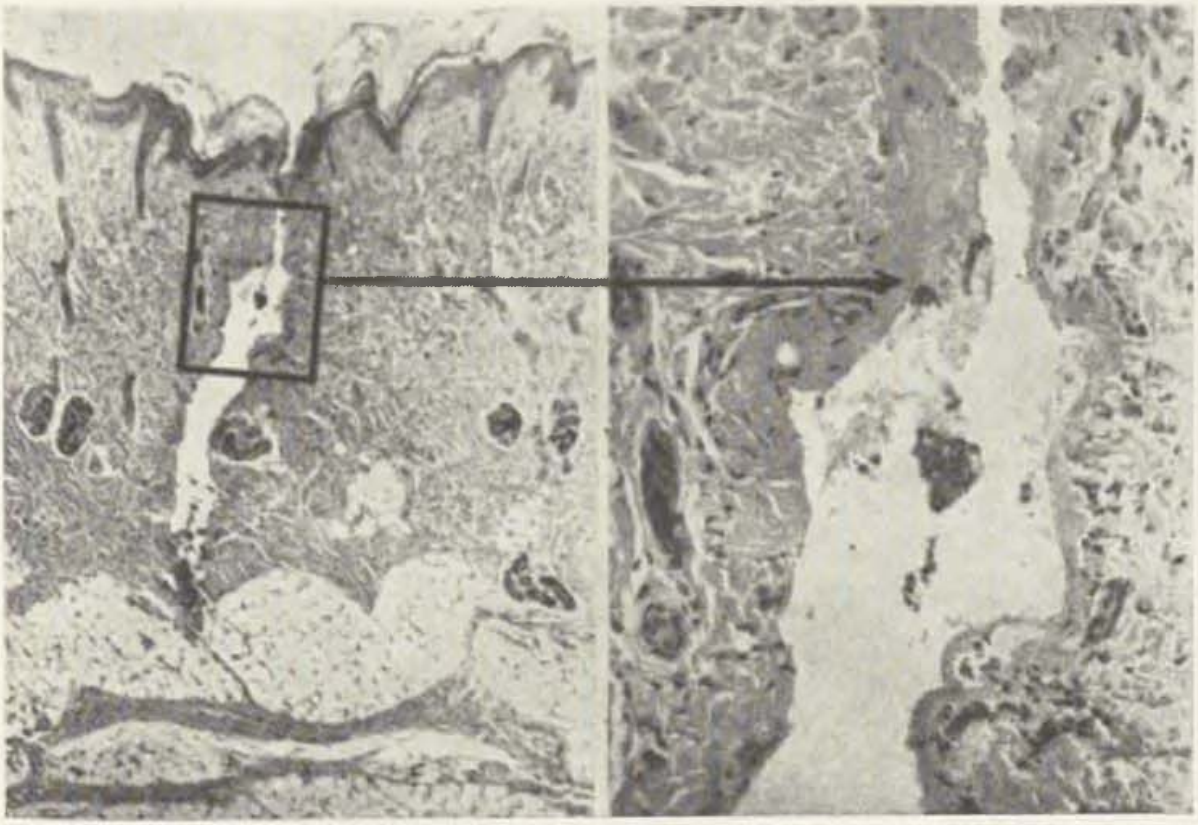

Fig. 7. Photomicrograph of skin incision one day postoperatively showing homogenous coagulum lining the wound walls.

In Fig. 7 the homogenous coagulum is seen lining the walls of the incision. This coagulum could be fibrin which has clotted as it exudes from the dermal walls, but we believe it is probably composed of the partially lysed ends of collagen fibres. The pH of wounds at this stage falls to as low as 6.4 in the depths of the incision (2, 3) and remains distinctly acid throughout

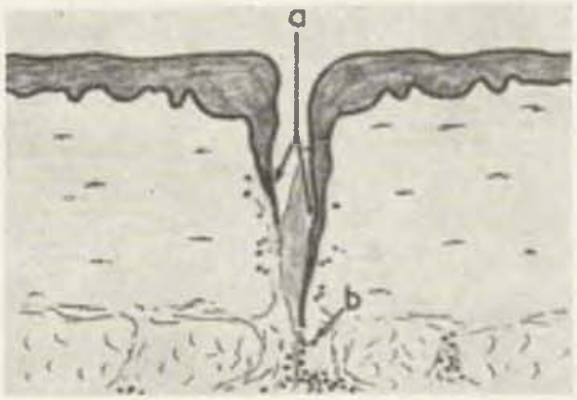

Fig. 8. Composite diagram of healing incisions from the period two to 14 days postoperatively. (a) Epidermis migrating down wound walls, and (b) phagocytes removing debris.

the first week or two of healing. The surface of the wound may be alkaline owing to evaporating carbonic acid. Collagen is partially soluble in weak acid and alkali. The altered wound pH, together with proteolytic enzymes released from degenerating and invading cells, is the most likely cause of what appears to be partial lysis of the dermal walls. Wound acidity is also thought to be partially responsible for the hyperemia, increased capillary permeability, and resulting protein exudation noted in the wound area (4).

Two to 14-Day Period

In our study the two to 14-day period of healing (Fig. 8) was characterized by epidermal migration into the incision, and by phagocytosis of clot and debris lying deep in the wound. Fig. 9, a histological section of an incision during this period, shows the migrating layer of epidermis advancing downward from the zone of epidermal thickening a short distance from the cut edge of epidermis. This thickening, also observed by others (4, 27), appears to be initiated by hypertrophy and continued by hyperplasia of the epidermal cells close to the cut edge. In this study, epidermal migration was observed to take place under clot and necrotic tissue, and on top of apparently healthy tissue as noted by previous investigators (7, 11). Edwards and Dunphy (9)

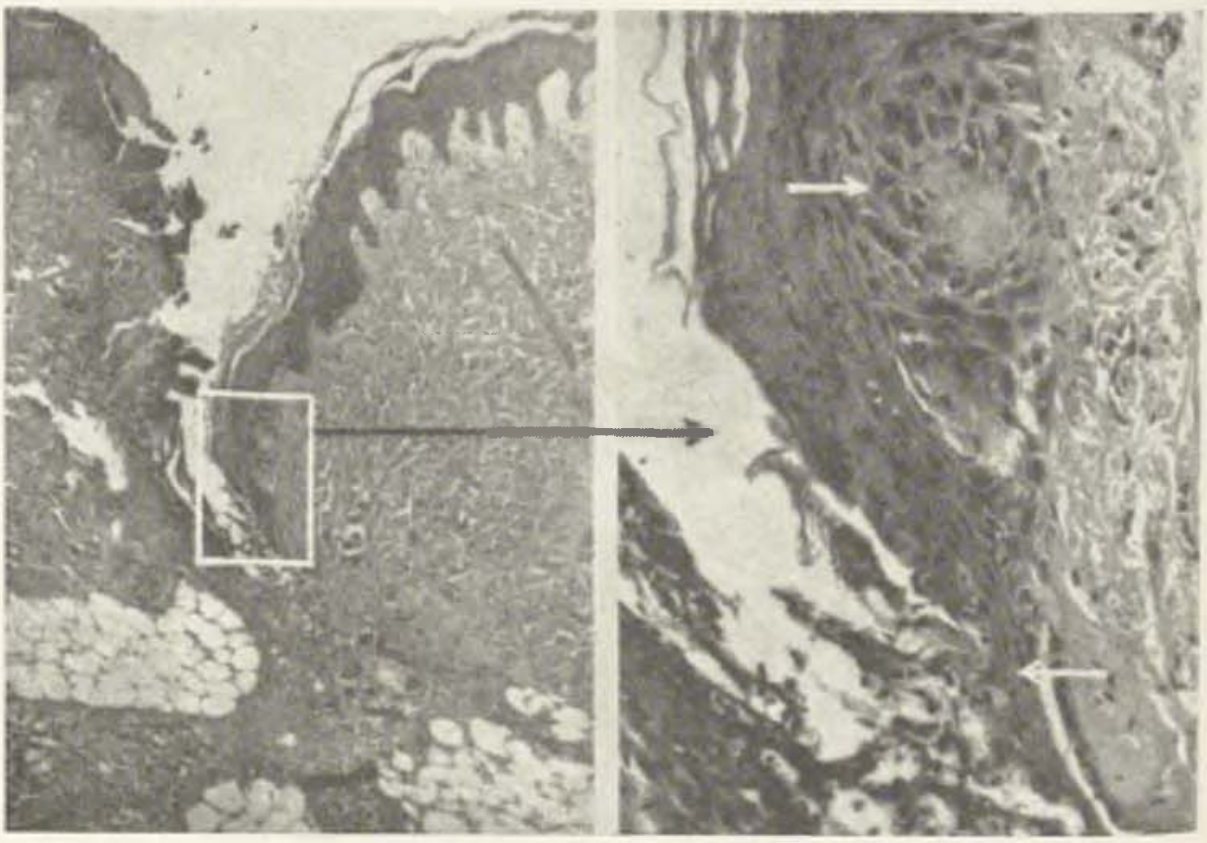

Fig. 9. Photomicrograph of skin incision three days postoperatively showing epidermal thickening at the wound margin (upper white arrow) and migration down wound walls under debris (lower white arrow).

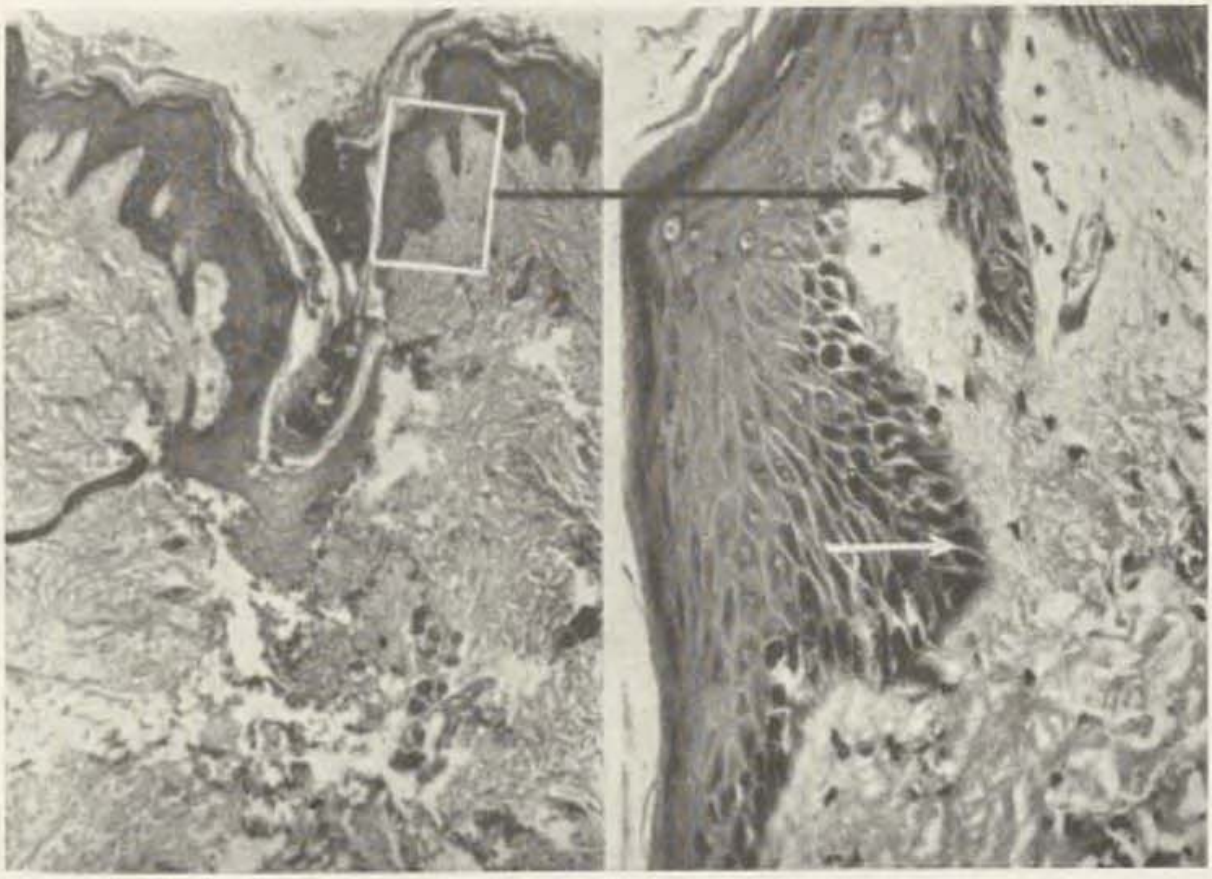

Fig. 10. Photomicrograph of skin incision 14 days postoperatively. Numerous mitoses are present in the basal layer of the epidermis (white arrow).

timed the onset of epidermal migration as a few hours after wounding, but we observed the first epidermal migration at about the second day postoperatively.

Studies with the electron microscope (32) have revealed that the cells of the central or prickle-cell layer of the epidermis are connected to each other and to the basal layer by minute tonofilaments. The basalcell layer is in turn

Fig. 11. Composite diagram of healing incision from the period 14 to 30 days postoperatively. (a) Epidermal proliferation into dermal and connective tissue defects, and (b) granulation tissue ridge.

connected by tonofilaments to dense rods anchored in the dermal membrane, superficial to the basement membrane. With so many interconnections it is hard to visualize how epidermal migration can take place, but Weiss (34) has demonstrated that following wounding in amphibian skin, similar connections are dissolved, thus allowing ameboid migration of individual epidermal cells into the defect. This has been verified by others (9, 19) who have also described elongation of the migrating cells and have suggested that the advancing cell border probably secretes a proteolytic enzyme which makes removal of obstructing clot and debris possible (5). The advancing epidermal cells are derived mainly from the prickle-cell layer (19). In the rabbit the epidermis of the ear migrates at about 3/10 mm. per day (24).

Fig. 10 shows another healing incision at this stage. The epidermal mitoses were most marked at the wound margins, and were most numerous about the time that epidermal continuity was established. This agrees with the findings of McMinn (27).

The three to six-day period immediately following wounding was previously referred to as the "lag period", because during this period there is no increase

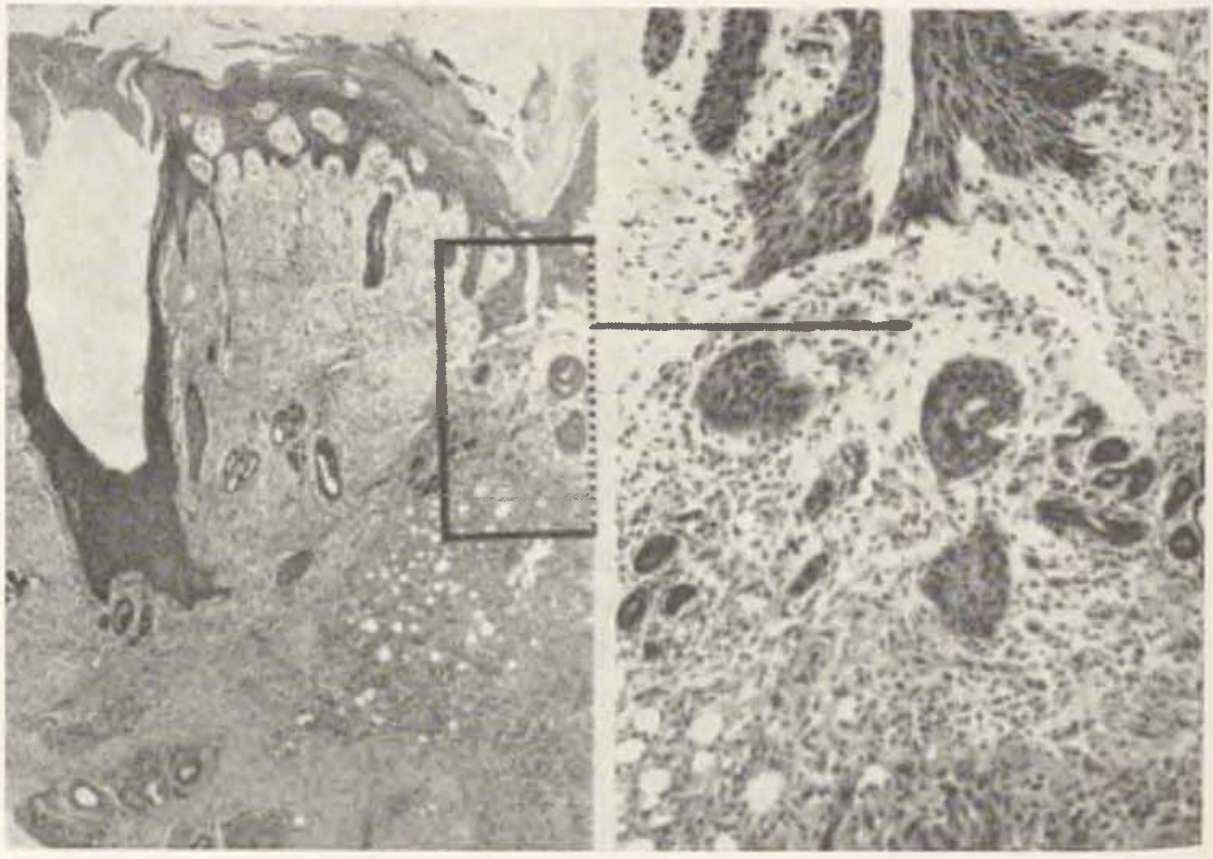

Fig. 12. Photomicrograph of a section taken 23 days postoperatively showing two incisions in the low-power view (left illustration). Only the right incision has penetrated the dermis to stimulate the subcutaneous fibroplasia which is seen magnified in the illustration on the right which has resulted in the isolation of several apparent epidermal islands (white arrow). We did not verify the existence of epidermal islands by using serial sections.

in the tensile strength of the wound (20, 21). It is now known that this is actually a period of great activity in the base of the wound. Therefore, it has been renamed the "preparatory period" by Dunphy (7). This period is characterized by cellular activity deep in the wound where macrophages, possibly originating from mononuclear blood cells (3, 28), phagocytose clot and tissue debris, and leukocytes remove bacteria.

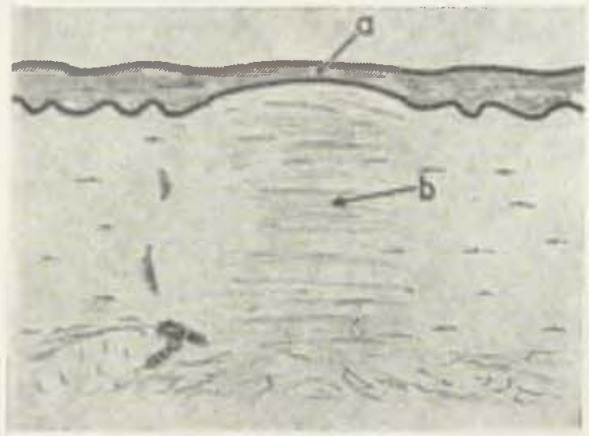

Fig. 13. Composite diagram of healing incisions from the period after 30 days postoperatively. (a) Raised thin epidermis, and (b) dense maturing scar.

Young fibroblasts appear during this period. Controversy exists as to the origin of these cells. A few are produced by the mitoses of surrounding normal fibroblasts, but many observers feel that the number of mitoses in the wound area is not sufficient to account for the great number of new fibroblasts which make their appearance in the wound during the first few days after incision. Other fibroblast sources have been postulated on the basis of some convincing evidence. They include the primitive cells surrounding capillaries and skin accessories (26, 29), mononuclear blood cells exuded into the wound (1, 14, 28), and macrophages (3). Whatever their origin, fibroblasts in this and other studies begin to appear during the first few days after wounding (31).

It is at this period that the first new collagen is formed in the wound. This is probably produced by fibroblasts as a fluid substance composed of polymers of tropocollagen which are dispersed into the intercellular matrix where they aggregate to form collagen fibrils (8, 12, 13, 22). These fibrils become progressively less soluble with age (30). Enough collagen fibres have been formed by the sixth postoperative day to increase the wound's tensible strength (2).

New mucopolysaccharide is also produced in the wound but this occurs much later than previously suggested by Dunphy and Udupa (8). Recent investigations (23) have demonstrated that chondroitin sulfate ground substance is not produced until about the ninth day after wounding. This agree with the finding that sulfate, S35, is not takeu up by the wound during the first few days after incision (16).

14 to 30-Day Period

Fig. 11 illustrates diagrammatically the healing skin wound 14 days to one month after incision. Healing at this stage is characterized by epidermal closure over the wound surface, epidermal penetration into dermal and connective tissue defects, and active fibroplasia from the base of the wound.

Fig. 14. Photomicrograph of skin incision 34 days postoperatively showing the epidermis in almost normal position and the early scar (white arrow) with its randomly arranged fibres.

Fibroplasia occurs in a ridge of granulation tissue composed of capillaries, fibroblasts and fine collagen fibres. This ridge originated from the subcutaneous tissues and wells up into the wound defect (7, 11, 19). Some subcutaneous fat cells appear to be trapped within it.

Fig. 12 shows a histological section in which two incisions have been made. The incision on the right has been carried through to the subcutaneous tissue stimulating a ridge of granulation tissue to well up as described above. The inadvertent incision on the left has not penetrated the dermis and has not stimulated the subcutaneous tissue. Consequently, fibroplasia is much less active in the left incision. The great majority of connective tissue originates from the subcutaneous fatty layer, while the dermis remains uninvolved. Gillman

et al. (11) also noted some fibroplasia originating in the papillary layer of the dermis, but our studies did not confirm this finding.

The epidermis recedes upwards as the connective tissue ridge wells up from below. In the process of this retreat some epidermal protrusions appear to become isolated and to form islands of epidermal cells surrounded by new connective tissue. Fig. 12 (right) shows a few such apparent islands which

Fig. 15. Photomicrograph of skin incision 140 days postoperatively showing the masses of collagen and amorphous elements present in dense mature scar (white arrow).

appeared on random sections. Although Gillman et al. have observed similar islands to remain for several months, they state that these isolated epidermal cells are eventually phagocytosed.

After the 30-Day Period

Fig. 13 is a diagram of a healing skin wound during the period from one month onward. Healing at this stage is characterized by a thin raised epidermis with trophic or absent epidermal pegs and a dense fibrous scar.

Fig. 14 shows a section through a healing incision early in this period. The ridge of granulation tissue has filled the wound space and forced the epidermis upwards to an almost normal position. Fig. 15 shows a section through a healing incision late in this period when the collagenic fibres have become oriented. Fig. 16 compares the early incomplete fibre orientation 34 days postoperatively, as in Fig. 14, with the later more complete fibre orientation 140 days postoperatively, as in Fig. 15.

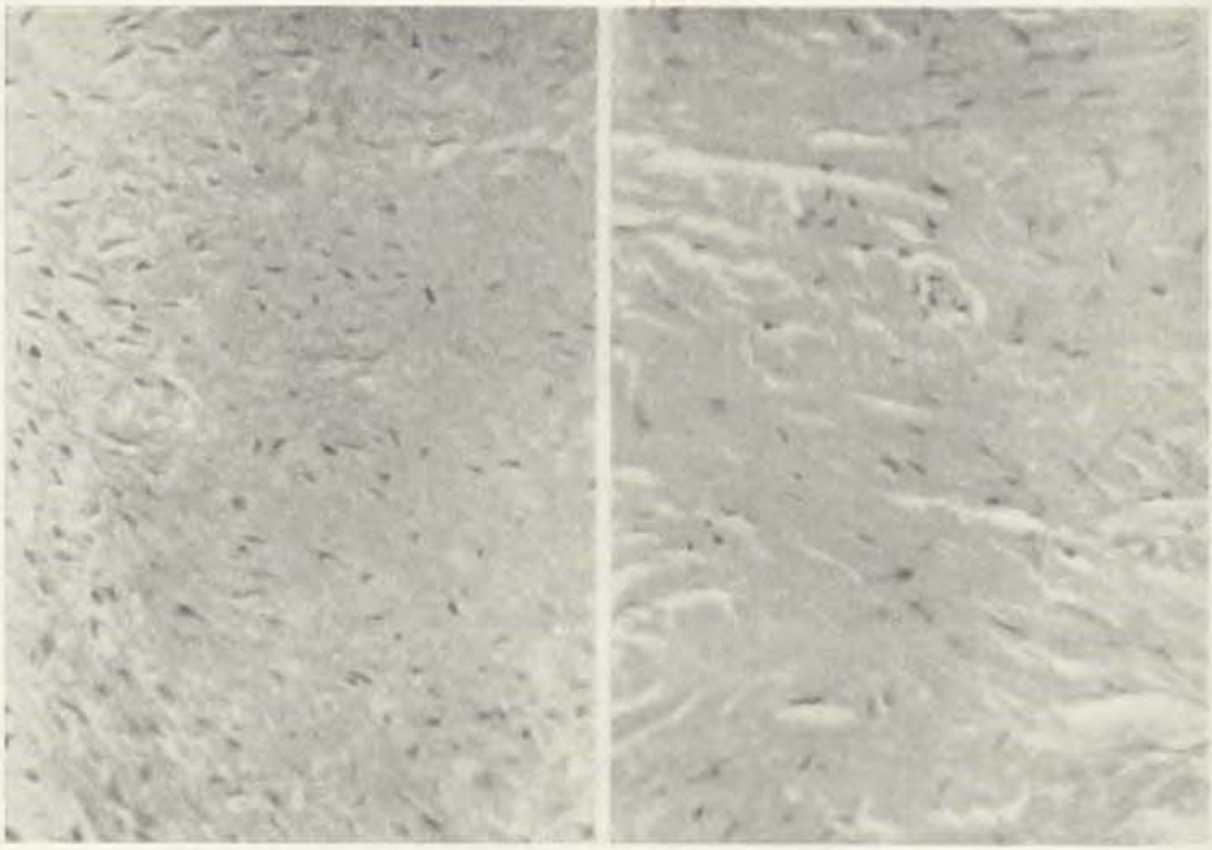

Fig. 16. Photomicrograph showing, on the left, the 34-day-old scar (delicate collagenic strands, cellular and disoriented) and on the right, the 140-day-old scar (coarse collagenic masses, less cellular and better oriented).

Gross (12) states that the collagen content of wound granulation tissue rapidly increases during the latter part of the first week after wounding, then decreases from about the seventh to the tenth postoperative day, only to begin a slow and very prolonged rise from about the tenth day onward. A comparison of the early and the mature scar reveals that the collagenic fibres and vessels, which at first are disoriented or lie parallel to the walls of the incision, eventually become oriented parallel to the skin surface. These fibres gradually increase in size for many months (18), and as they become larger the scar contracts, often deforming surrounding tissues as it does so. This phenomenon is properly referred to as "cicatrization" to distinguish in from "contraction" which is the phenomenon largely responsible for the spontaneous closure of excised and gaping wounds (33). Cicatrization has been ascribed to shortening of the collagen fibres (19), but fibre shortening has never been proved.

Capillaries and cells gradually disappear from the scar as its collagen content increases. This is thought to cause the slightly raised pink scar to become flat and white.

Skin appendages such as hair follicles and sweat glands are usually absent from the scar. If present they are frequently deformed. Gillman (10) states that epidermis can regenerate skin appendages, but we have not observed this.

The loss of skin appendages, cells and capillaries may be due to increasing pressure within the scar as it matures. The epidermis does not become normal even after it returns to an almost normal position. Its thickness is often reduced and its epidermal pegs are usually atrophic, and may be entirely absent, as shown in Fig 17.

Fig. 17. Photomicrograph of a skin incision 51 days postoperatively showing elevated epidermis without epidermal pegs.

The results obtained in this study of thin skin healing agree in general with those of Gillman et al. (11) and Dunphy (7) with the chief exception that healing appears to have been slower in the subjects observed by us. The fact that our subjects were all children should have increased the healing rate (35). The retarding influences of a variety of systematic diseases, edema around the cutdown site, and a peripheral site if incision, appear to have delayed healing beyond that accepted as normal by other workers.

CONCLUSIONS

Scar formation is to be avoided since a wound with heavy scarring takes longer to heal, is unsightly, produces deformity and contraction, and is more liable to break down that the surrounding normal skin. Scar formation may be minimized by careful apposition of the dermis to avoid the formation of dead space which must be filled by the granulation-tissue ridge from the

subcutaneous zone. Skin sutures should be placed to accomplish this and to counteract the spontaneous inversion of the cut skin edges. They should penetrate only skin and should not include subcutaneous tissues, which they will pull upwards into the dermal defect. Subcutaneous sutures should be tied to produce just enough tension to hold subcutaneous tissues in apposition without producing a ridge which will push upwards into the dermal defect.

Technique should always be gentle, since irritation or hemorrhage by increasing inflammation increase granulation tissue and leads to increased scar formulation.

SUMMARY

The healing of human incised wounds in thin skin was studied, using as experimental material cutdown incisions over the long saphenous vein at the ankle taken from autopsy cases.

The healing process was characterized by the following sequence of events: Exudation of protein and cells; epidermal migration under clot and debris to line the wound walls; granulation tissue, resulting from subcutaneous fibroplasia, welling up to fill the wound space and forcing the new pidermis upward, and scar maturation with collagenic fibre orientation and epidermal thinning.

This sequence of events was found to correspond closely to that described by Gillman et al. (11) with the exception that healing took place more slowly in this study.

The literature on skin healing has been reviewed briefly.

RÉSUMÉ

La guérison de la peau

W. K. Lindsay, J. R. Birch

De nombreuses études ont déjà été faites sur la cicatrisation des plaies cutanées, mais la plupart d'entre elles relèvent d'expérimentation sur l'animal. Après avoir passé en revue la littérature sur le sujet, les auteurs exposent les résultats d'études personnelles. Leur matériel consiste en 27 spécimens prélevés à l'autopsie sur des enfants qui avaient reçu des injections par cathéter dans la veine saphène. La région d'insertion du cathéter fut soigneusement excisée, fixée à la formaline et étudiée histologiquement. Les avantages de ce matériel étaient son homogénéité, provenant d'une même localisation et de sujets dont les âges variaient entre un jour et trois ans et demi. Les inconvénients possibles étaient l'infection et l'état pathologique; cependant les phénomènes d'infection étaient réduits à un minimum et le facteur morbide ne semble pas avoir joué un rôle important si ce n'est en produisant un ralentissement des phénomènes. Le processus de cicatrisation peut être subdivisé en diverses périodes: (1) Période entre zéro et six heures. — On note à ce moment une inversion des bords de la plaie et des foyers hémorragiques accompagnes d'une exsudation de plasma et de lymphe; tout ceci tend à former un réseau fibrineux. (2) Entre six heures et deux jours, on assiste à un épaississement progressif de l'épiderme; des cellules inflammatoires envahissent le terrain et tendent à détruire la caillot. (3) Dans la période située entre deux et 14 jours, on note une migration épidermique dans l'incision et une

accélération des phénomènes de phagocytose au sein du caillot; la migration des cellules épidermiques a été étudiée au microscope électronique: on sait que ces cellules sont fortement fixées les unes aux autres par un système complexe de tonofibrilles et il semble qu'à un certain moment, une dissolution temporaire de ces tonofibrilles se fasse, permettant ainsi aux cellules de se déplacer par mouvements amiboïdes. Dans la profondeur, on voit une prolifération de jeunes cellules conjonctives. (4) Période entre 14 et 30 jours. Ce stade est caractérisé par la fermeture de la déhiscence épidermique et par une pénétration de l'épiderme en profondeur; de plus, le tissu dermique s'organise. (5) Période après 30 jours. Il se fait une orientation dans l'arrangement des fibres collagènes dermiques et l'épiderme réduit son épaisseur.

ZUSAMMENFASSUNG

Wundheilung von Hautwunden

W. K. Lindsay, J. R. Birch

Es wurde die Heilung von Inzisionen in dünner Haut untersucht, wobei Hautinzisionen über der Vena saphena in Knöchelhöhe anhand von Sektionsmaterial studiert wurde.

Der Heilungsprozess war durch folgende sukzessive Vorgänge charakterisiert: Exsudation von eiweisshaltiger Flüssigkeit und Zellen; epidermale Migration unter dem Koagulum und Detritus, die Wundränder säumend; subkutane Fibroplasie produziert Granulationsgewebe, das die Wunde ausfüllt und die neue Epidermis höher verlagert; schliesslich Narbenreifung mit Ausrichten der kollagenen Fasern und Verdünnen der Epidermis.

Diese Reihenfolge der Heilungsvorgänge entspricht genau derjenigen, die Gillman und Mitarbeiter beschrieben haben, mit der Ausnahme, dass die Heilung in unseren Fällen langsamer verlief.

Es wurde eine kurze Übersicht über die Literatur inbezug auf Heilung von Hautwunden gegeben.

RESUMEN

La curación en la piel delgada

W. K. Lindsay, J. R. Birch

Fué estudiada la curación de las heridas humanas incisas, en la piel delgada, usando como material experimental incisiones de cortes bajos sobre la vena saphena magna en el tobillo, tomadas de casos de autopsia.

El proceso de curación fué caracterizado por la siguiente serie de acontecimientos: exsudación de proteínas y de células; la migración epidérmica debajo del coágulo y de los desechos hacia las paredes de la herida; el tejido de granulación, resultante de la fibroplasia subcutánea, brotando hacia arriba para llenar hasta el borde, el espacio de la herida y forzando la nueva epidermis con dirección hacia arriba, y la maduración de la cicatriz con la orientación de las fibras colágenas y el adelgazamiento de la epidermis.

Esta serie de acontecimientos fué averiguada para corresponder estrechamente a lo descrito por Gillman y sus seguidores, con la excepción de que la curación, en este estudio, fué más lenta.

La literatura de la curación de la piel, ha sido analizada brevemente.

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The authors wish to express their gratitude to Dr. Sylvia H. Bensley, University of Toronto, for her assistance in histological interpretation; to Dr. V. W. Krause, Pathology Department, the Hospital for Sick Children, for providing the material for study, and to Miss Lilian Harrison for preparing the microscopic sections.

This project was assisted by funds allocated by the Province of Ontario under the National Health Program of the Department of National Health and Welfare Ottawa, Ont. Grant Number 605—13—42.

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THE EFFECTS OF AN ANTIHISTAMINE (PROMETHAZINE) ON THE REACTION OF TENDONS TO TRAUMA

(A histological study)

F. G. WALKER, S. H. BENSLEY, W. K. LINDSAY

ABSTRACT

As a part of longterm work on the healing of flexor tendons, the effects of an antihistamine-antiserotonin compound (promethazine) were studied in 109 chickens. A form of standard surgical trauma to the intact flexor profundus tendon was devised, and the results of several different methods of administration of promethazine were compared. These studies indicate that minimal surgical trauma applied to an intact flexor tendon produces a marked reaction consisting of a degenerative phase, a phase of dedifferentation, and a phase of regeneration and maturation. The histological studies reveal that such highly specialized intercellular substance as collagenic fibers and bundles, though non-living, are not inert and permanent structures. They can break down, completely disappear, and be reformed as a result of the reaction to trauma of specialized tenocytes and undifferentiated mesenchymal cells of the tendon and epitenon. The effects of an antihistamine (promethazine) are to prolong the phase of degeneration and loss of differentiation and, upon withdrawal, to accelerate the phase of growth and maturation. These effects vary with the dose and method of administration of promethazine.

INTRODUCTION

The effects of antihistamine drugs on many physiological and pathological processes have been studied, but there have been few reports on the effects of such drugs on fibroplasia. Berman et al. (1) found that methapyrilene HCl [Histadyl (R)] limited the amount of fibroplasia in experimental soft tissue wounds produced by severe or continued trauma. This effect was not observed in incised wounds where there had been minimal damage to cells. Hormia and Hormia (2) found that wound healing in the rat was slowed slightly by the use of chlorpromazine [Largactil (R)]; this effect was not statistically significant. Weil et al. (3) reported that pyrrobutamine [Pyronil (R)] retarded the

healing of corneal incisions in the rabbit. Conversely, Baldridge [4] reported that pyrrobutamine produced a significant increase in the rate of healing of wounds in the guinea pig. There have been no reports of the effects of such drugs on tendon healing.

As part of a longterm work on the healing of flexor tendons [5], the effects of an antihistamine on the healing of traumatized chicken tendons

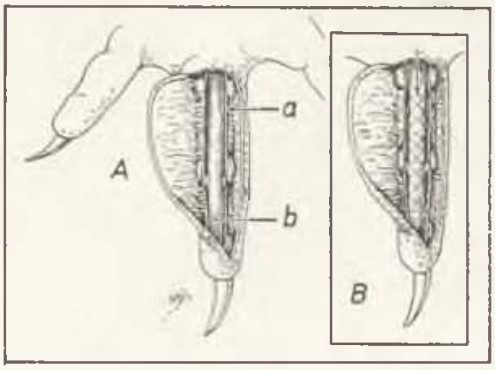

Fig. 1. [A] Control procedure, flexor sheath and flexor sublimis tendon excised. [B] Standard surgical trauma: 0000 silk inserted 18 times into intact flexor profundus tendon. a — Edge of excised sheath, b — flexor digitorum profundus tendon.

were studied. Promethazine HCl [Phenergan [R]] was chosen for this experiment because it possesses potent antihistamine activity [6]. It also antagonizes the action of serotonin [7]. Histological aspects of this study are presented.

METHODS

One hundred and nine female chickens, unselected as to breed, 6 to 8 months of age, and weighing from 2300 to 3400 g were used throughout the experiment. They were fed on a standard diet consisting of scratch grain, egg pellets, egg mash, grit, and oyster shell, and were given water and libitum.

Preoperatively, chickens were starved for 18 hours. Sedation was obtained with pentobarbital sodium [Nembutal [R]] I.M. 20 mg/kg. Regional anaesthesia was obtained by using lidocaine 1 % [Xylocaine [R]] as a femoral and sciatic nerve block. A tourniquet was applied to each foot prior to the surgical procedure.

A standard operation was carried out to obtain a surgical control. Under surgically sterile conditions, using an L"shaped incision, a palmar skin flap was reflected from the center toe, the flexor sheath was excised throughout the length of the toe, and the flexor sublimis tendon was excised from its insertion to as high in the foot as could be reached from the surgical exposure. Skin edges were then apposed with interrupted 0000 silk sutures.

Standard surgical trauma was produced using a similar surgical exposure. In addition, a 0000 silk suture on a No. 14 curved cutting needle with a French eye was inserted 18 times into the intact flexor profundus tendon [Fig. 1].

The suture was then removed prior to apposing skin edges. The tendon was held as necessary with toothed Adson forceps (Ingram and Bell, Catalogue No. 533).

A combination of the insertion and removal of the silk suture plus the handling of the tendon with toothed Adson forceps produced a zone of standard tendon trauma 1 cm. on each side of the attachment of the vinculum longum, free

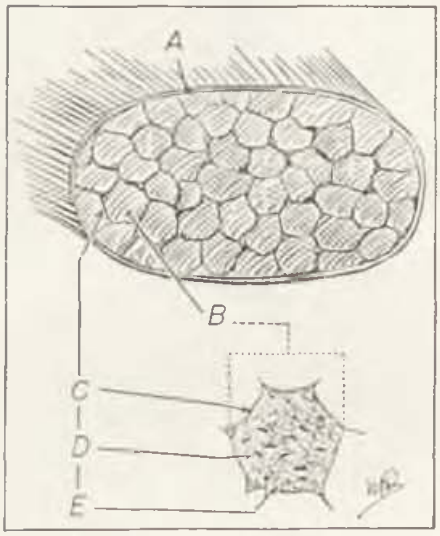

Fig. 2. Schematic cross section of normal flexor tendon. A — epitenon, B — tendon bundle, C — endotenon, D — tenocyte, E — collagenic fibre.

from possible foreign body reaction. The surgical control procedure parallelled the standard surgical trauma procedure in all respects except for the administration of trauma to the intact flexor profundus tendon.

Both feet in all chickens were subjected to a surgical procedure. In certain groups the tendon of the long toe on the other foot was divided transversely and repaired with 000000 stainless steel wire. The results from this experiment are described elsewhere (8).

Postoperatively, both feet were immobilized in plaster of Paris casts for 3 weeks, or until the experiments was terminated. After removal of the casts the chickens were allowed to walk freely, and were perched for 4 to 6 hours per day on a 1 in. diameter rod. Skin sutures were removed 4½ weeks postoperatively.

The effects of several methods of drug administration were compared as follows.

Group I: Surgical control. No drug was administered.

Group II. Standard trauma. No drug was administered.

Group III: Standard trauma. Promethazine 15 mg (0.6 cc) was instilled around the tendon immediately after the skin closure, using a No. 27 hypodermic needle inserted between the skin edges.

Tab. 1. Average cell counts (10 fields) of all nuclei except those

	Hours	Weeks
	48	1
Group I: (Surg. cont.)	30.2 (20—40)	30.9 (20—41)
Group II: (Standard trauma)	30.2 (18—46)	72.5 (40—130)
Group III: (Promethazine top (15 mg))	30.5 (24—38)	27.4 (10—48)
Group IV: (Promethazine I.M. 1 day (300 mg))	28.2 (19—41)	31.8 (21—44)
Group V: (Promethazine I. M. 1 week (350 mg))	30.9 (18—42)	24.6 (18—42)
Group VI: (Promethazine top. (15 mg) promethazine I.M. 1 week (350 mg))	32.6 (24—48)	19.9 (10—28)
Group VII: (Promethazine I.M. 1 month (1800 mg))	30.5 (21—37)	31.2 (21—44)
Group VIII: (Promethazine top. (15 mg) promethazine I.M. 1 month (1800 mg))	33.7 (29—52)	30.2 (20—38)

Group IV: Standard trauma. Promethazine I.M., 100 mg, was administered into the pectoral muscles at 8 a.m., 12 noon, and 4 p.m. on the day of operation only. Operation was carried out in the early afternoon.

Group V: Standard trauma. Promethazine I.M. 25 mg b.i.d. was administered into the pectoral muscles for 7 days postoperatively.

Group VI: Standard trauma. Promethazine I.M. 25 mg b.i.d. was administered into the pectoral muscles for 7 days postoperatively. Promethazine 15 mg (0.6 cc) was instilled around the tendon immediately after skin closure, as in group III.

Group VII: Standard trauma. Promethazine I.M. 100 mg was administered three times on the day of operation in a manner similar to group IV. Promethazine I.N. 25 mg b.i.d. was given for 30 days postoperatively.

Group VIII: Standard trauma. Promethazine I. M. 100 mg was administered three times on the day of operation and promethazine I. M. 25 mg b.i.d. was given for 30 days postoperatively (i.e. the same regimen as group VII). Promethazine 15 mg (0.6 cc) was instilled around the tendon immediately after the skin closure, as in group III.

This enabled us to study the effects of standard surgical trauma alone (group II), and compare the effect on the reaction to standard surgical trauma of topical promethazine (group III), early administration of prometha-

in vessels and vessel walls. Range is given in parentheses

Weeks				
2	3	5	8	16
34.0 (32—52)	28.1 (18—40)	48.1 (36—64)	32.8 (18—52)	64.1 (44—80)
120.8 (85—215)	87.9 (16—164)	216.8 (144—256)	190.8 (140—240)	170.1 (140—204)
29.3 (16—40)	24.1 (10—43)	151.3 (59—214)	113.8 (88—144)	52.0 (15—77)
82.4 (62—115)	29.7 (20—44)	85.6 (65—151)	327.0 (160—400)	147.2 (90—192)
66.3 (52—99)	71.2 (44—120)	93.9 (51—120)	335.6 (318—352)	288.1 (230—402)
26.0 (8—43)	33.9 (16—60)	4.7 (0—23)	133.1 (89—188)	61.0 (33—82)
19.4 (9—36)	18.5 (15—21)	222.8 (138—278)	130.0 (104—158)	42.2 (15—68)
37.6 (28—45)	5.3 (2—11)	294.4 (256—328)	274.8 (200—452)	223.5 (194—282)

zine (group IV), two ranges of prolonged administration (groups V and VII), and combinations of these (groups VI, VII, and VIII).

In groups VII and VIII toxic levels of the drug were reached. Many of the chickens in these groups became lethargic, developed diarrhoea, lost weight, and developed pressure sores over the sternum and in the ankle creases.

Birds were killed by cervical dislocation at intervals of 48 hours, 1, 2, 3, 5, 8, and 16 weeks in all groups. At certain time intervals two or more birds were sacrificed. This was necessitated by illness of the bird and occurred mainly in groups VII and VIII. Most time intervals were represented by a single specimen. The feet were amputed and fixed in 10% formalin. Autopsy dissection of the toe subjected to operation was carried out and a segment of the tendon extending 1 cm on each side of the cinculum longum was removed. This segment was embedded in paraffin and sectioned transversely through the area of attachment of the vinculum longum. All sections were cut at five microns and stained routinely with hematoxylin and eosin. To facilitate comparison, photomicrographs (X64) were taken of all sections using Ectachrome EF 135 film.

Cell counts were carried out under oil immersion using a 10 X ocular and a 90 X objective. In each section studied, all nuclei, except those in vessels and vessels walls, in 10 fields within the tendon proper were counted.

The largest and smallest collagenic fibers were selected in the tendon in each section studied and the largest and smallest cross-sectional diameter of each was measured in microns using an ocular micrometer. The average of these two diameters of each fiber was used to represent the size of the fiber.

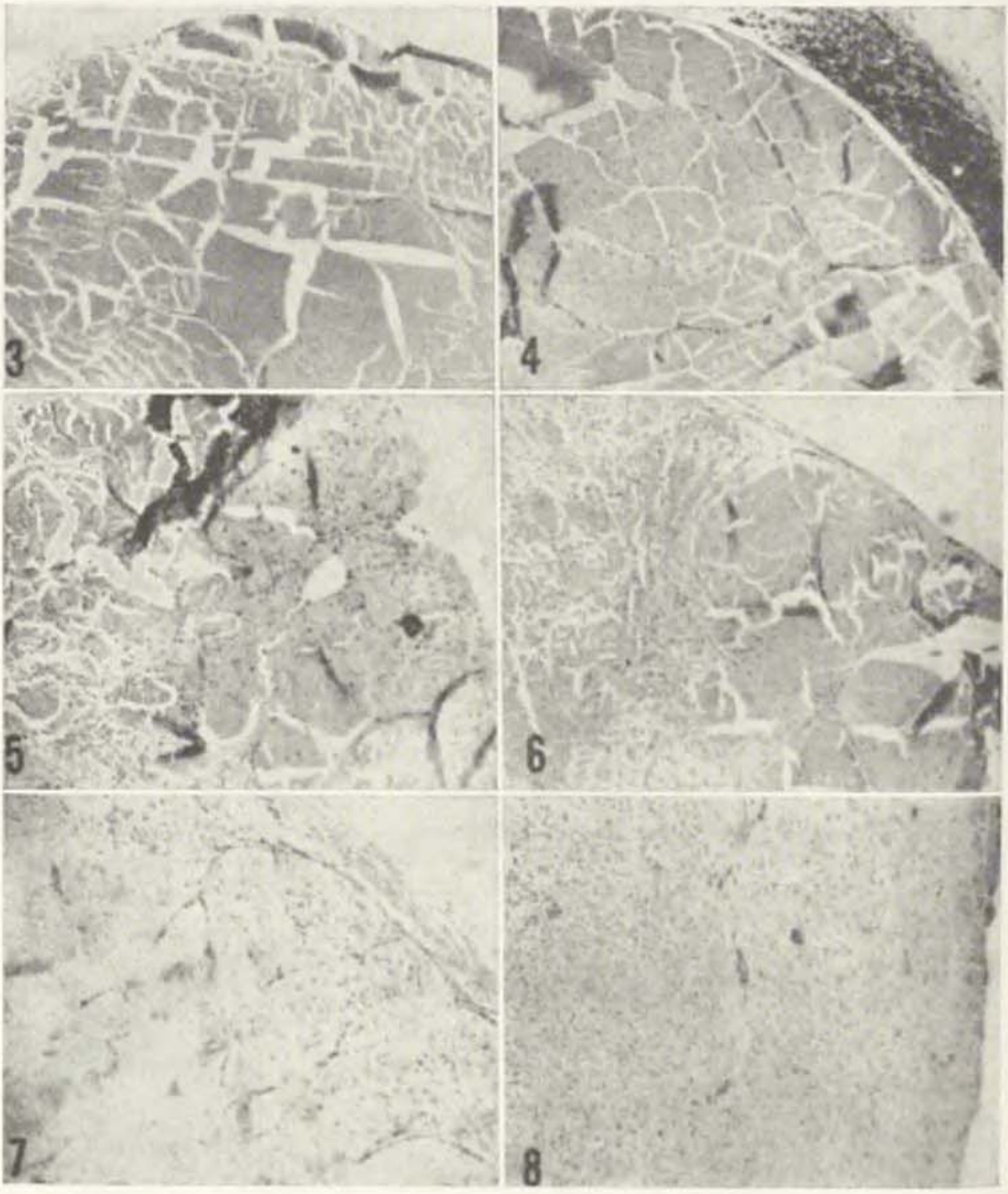

Fig. 3. Tendon cross section, group I, 2 weeks, X 256. — Fig. 4. Tendon cross section, group II, 48 hours, X 256. — Fig. 5. Tendon cross section, group II, 1 week, X 256. — Fig. 6. Tendon cross section, group II, 2 weeks, X 256. — Fig. 7. Tendon cross section, group II, 3 weeks, X 256. — Fig. 8. Tendon cross section, group II, 5 weeks, X 256.

OBSERVATION

Terminology

The microscopic anatomy of the normal flexor tendon is shown schematically (Fig. 2). The epitenon is a cellular layer completely enveloping the tendon. The endotenon is continuous with it and projects through the tendon substance, dividing it into tendon bundles. Like the epitenon, the endotenon is moderately cellular. It contains an occasional blood vessel. Tendon bundles are composed of collagenic fibers and fibrocytes which may be termed tenocytes. Mature collagenic fibers have regular outlines, and the tenocytes have little visible cytoplasm.

Group I: (Surgical Control, No Drug)

The microscopic anatomy of sections in the surgical control group is essentially normal. The epitenon is two or three cells in thickness; the endotenon is conspicuous, moderately cellular, and contains some vascular channels. The tendon bundles are well outlined, and contain collagenic fibers which have a regular border. Tenocytes are seen uniformly throughout the tendon. They have little visible cytoplasm. A typical section, taken 2 weeks after surgery, is illustrated (Fig. 3).

Average cell counts at all intervals studied after surgery are relatively constant (Table I), as are average diameters of largest and smallest collagenic fibers (Table II).

Group II: (Standard Trauma, No Drug)

Forty-eight hours (Fig. 4). The epitenon is thin. Immediately outside it is seen an acute inflammatory exudate. The endotenon is conspicuous, cellular, and vascular. Tendon bundles are well outlined. Many collagenic fibers are larger than those in the surgical control group; some are very small; most are frayed, i.e. the fiber borders are irregular and frond-like. This is more obvious with low illumination. Total cellularity in the cross-sectional area of the tendon proper is not increased. In the area of the largest fibers, there is a patchy loss of nuclei.

One week (Fig. 5). The epitenon is thick and cellular. The endotenon, which is poorly defined, shows increased vascularity and cellularity. Tendon bundles are partially replaced by cellular tissue. Collagenic fibers are frayed and small, although in some areas they are of normal diameter. The total number of nuclei is slightly increased.

Two weeks (Fig. 6). The epitenon is relatively thin but is quite cellular and very vascular, as is the endotenon. Many of the tendon bundles are replaced by cellular tissue. A very few areas show typical bundles. There are numerous nuclei in these areas. Almost all of the collagenic fibers are small; where typical bundles are seen they are of normal size. All fibers are frayed.

Three weeks (Fig. 7). The epitenon is several cells in thickness, and is moderately cellular and vascular, as is the endotenon, which is conspicuous. Most tendon bundles are distinct but show a marked increase in the number

of nuclei contained within them. Mitoses are seen in these areas. In some areas nuclei are not increased in number; collagenic fibers in these areas are of normal size. Where nuclei are increased, fibers are small. All fibers are frayed.

Five weeks (Fig. 8). The epitenon is not seen as an entity. The endotenon where recognizable is thick and cellular. It demarcates very cellular areas which are not recognizable as tendon bundles. The entire tendon is very vascular. Almost all of the collagenic fibers are very small and frayed, while a very few normal-sized fibers are seen. These, too, are frayed.

Eight weeks (Fig 9). A thin layer of cells, suggestive of a primitive epitenon, surrounds the tendon. Tendon bundles and tibers are completely replaced by cellular tissue. Within this, near the center of the tendon, there is a small area of fine collagenic fibers which stain more deeply and have a smooth outline. Associated with these fibers are cells with hyperchromatic nuclei and visible cytoplasm which envelops a fiber.

Sixteen weeks (Fig. 10). The epitenon and endotenon are cellular and vascular. The tendon bundles are small, as are most of the collagenic fibers. The fibers are fairly well oriented linearly, and have a smooth border. The total number of nuclei themselves is small.

Cellularity and Fiber Size: (Tab. 1, Tab. 2).

Average cellularity, as measured by cell counts taken in the cross-sectional area of the tendon proper, increases with time to a maximum at the fifth week. It then decreases slightly until the 16th week.

The average diameter of the largest collagenic fiber is increased 48 hours after standard trauma. It then falls steadily until the fifth week, to return to normal levels or slightly higher by 16 weeks. Very small fibers are seen as early as 48 hours after operation and fibers smaller than normal are seen at all times studied after standard surgical trauma.

Group III: (Standard Trauma: Promethazine 15 mg Topically)

Forty-eight hours, 1, 2, and 3 weeks after standard surgical trauma, the general architectural pattern of the tendons studied varies little from normal. Collagenic fibers are generally larger than normal and have frayed borders. Five and eight weeks after trauma, the histology is similar to that seen in group II at 2 weeks. Sixteen weeks after operation, the general architectural pattern is similar to that of group II at the same time interval. Cellularity is less, and fiber size is generally greater. Very lew frayed fibers are seen at all intervals studied.

Group IV: (Standard Trauma; Promethazine I. M. 300 mg Given on Day of Operation)

Forty-eight hours, 1 and 3 weeks after standard trauma, the architectural pattern of the tendon is normal, but collagenic fibers are generally large and frayed. No very small fibers are seen before 2 weeks. At 2 and 5 weeks after trauma, the histology is similar to that seen in group II at 1 week (Fig. 11). At 8 weeks, the microscopic anatomy is similar to that in group II, 8 weeks.

but many more small well-stained fibers are seen enveloped by cytoplasm and accompanied by hyperchromatic nuclei (Fig. 12). The picture seen at 16 weeks is similar to that of group II, 16 weeks.

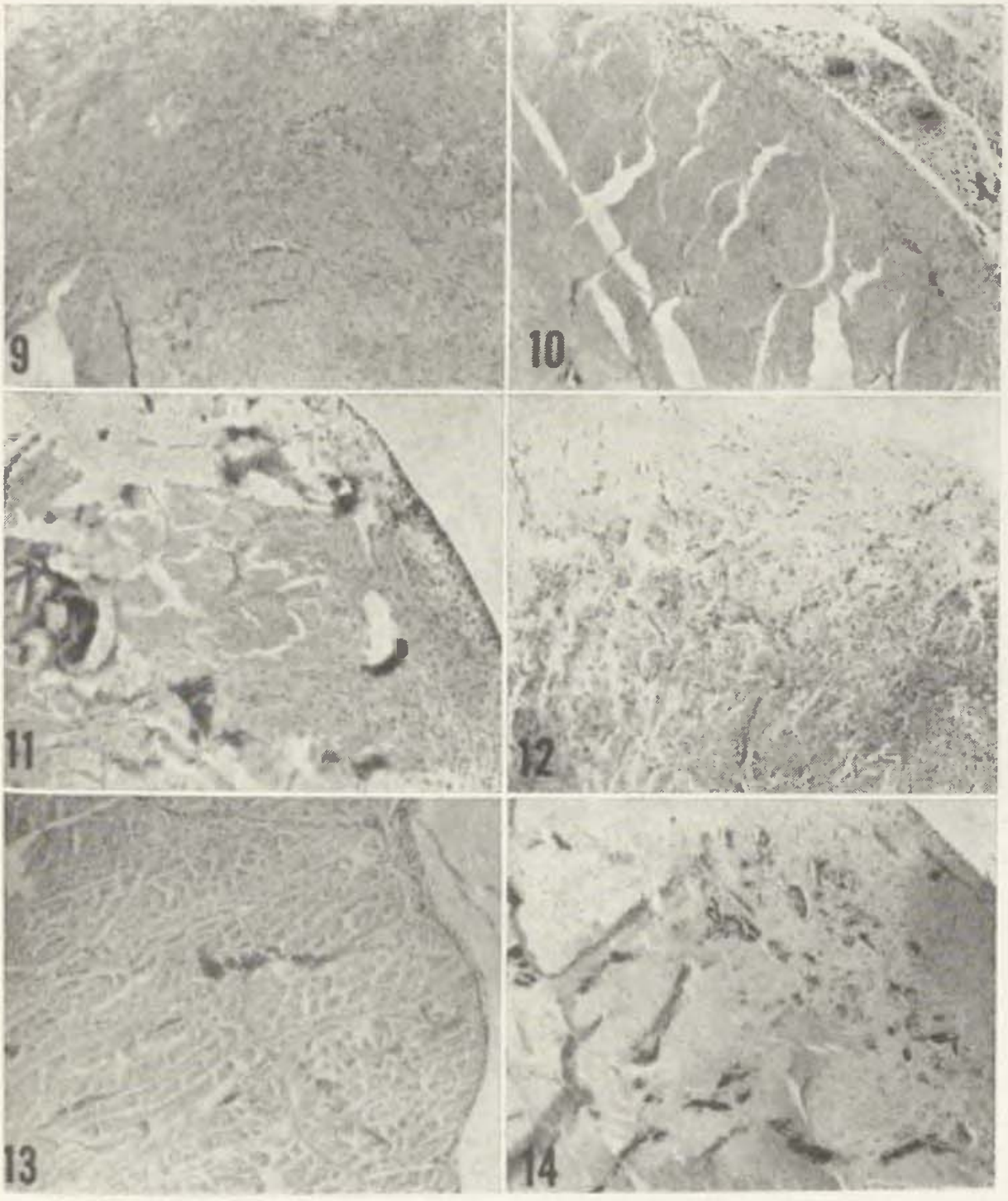

Fig. 9. Tendon cross section, group II, 8 weeks, X 256. — Fig. 10. Tendon cross section, group II, 16 weeks, X 256. — Fig. 11. Tendon cross section, group IV, 5 weeks, X 256. — Fig. 12. Tendon cross section, group IV, 8 weeks, X 256. — Fig. 13. Tendon cross section, group V, 3 weeks, X 256. — Fig. 14. Tendon cross section, group VII, 5 weeks, X 256.

Tab. 2. Average collagenic

	Hours,		Weeks		
	48		1		2
	L*	S*	L	S	L
Group I: (Surg. cont.)	2.70	0.90	2.20	1.00	2.37
Group II: (Standard trauma)	4.55	0.47	1.65	0.27	2.87
Group III: (Promethazine top. (15 mg))	3.30	0.57	4.47	0.45	4.15
Group IV: (Promethazine I.M. 1 day (300 mg))	2.65	1.70	4.25	1.85	3.60
Group V: (Promethazine I.M. 1 week (350 mg))	2.07	0.45	2.37	0.40	2.85
Group VI: (Promethazine top. (15 mg); promethazine I.M. 1 week (350 mg))	3.25	0.60	2.70	0.45	3.85
Group VII: (Promethazine I.M. 1 month (1800 mg))	2.77	1.52	4.75	1.67	8.30
Group VIII: (Promethazine top. (15 mg); promethazine I.M. 1 month (1800 mg))	3.27	0.57	3.67	0.45	2.90

L = largest; S = smallest.

Group V: (Standard Trauma; Promethazine I.M. 350 mg over 7-Day Period)

The general architectural pattern is near normal for the first 5 weeks after standard trauma (Fig. 13). Fiber size and total cellularity in the cross-sectional area of the tendon proper is normal for the first 3 weeks; most fibers are frayed; very small fibers are seen early. Fibers are small and markedly frayed at 5 weeks, and cellularity is increased. Eight weeks after operation, the tendon resembles that in group II, 5 weeks. The architecture is completely obliterated by very cellular, vascular tissue. The picture at the 16-week interval is similar to that of group II, 16 weeks, although cellularity is greater.

Group VI: (Standard Trauma; Promethazine I.M. 350 mg over 7-Day Period; Promethazine 15 mg Topically)

Forty-eight hours to 3 weeks after trauma, tendons show a normal architectural pattern. Cellularity is normal; fibers are generally large and frayed. Five weeks after trauma, there is a marked decrease in the total number of nuclei seen within the tendon proper, although the architectural pattern is normal. The sections at 8 weeks are similar to those of group II at 2 weeks. The epitenon and endotenon are nearly normal, but many tendon bundles are replaced by cellular, vascular tissue. Sixteen weeks after trauma the microscopic anatomy seen in group II at 16 weeks is repeated, although total cellularity is less. At all times studied small fibers are seen.

fiber size in microns

Weeks								
2	3		5		8		16	
S	L	S	L	S	L	S	L	S
1.13	2.40	1.20	3.55	1.00	2.95	0.97	2.05	1.22
0.40	2.65	0.40	1.50	0.37	0.10	5.34	1.55	0.65
0.60	3.85	0.32	1.45	0.32	1.25	0.40	5.25	0.55
0.60	2.50	0.50	3.20	0.47	2.75	0.40	2.90	0.27
0.35	2.67	0.22	2.00	0.25	0.80	0.37	2.65	0.30
0.35	4.40	0.40	3.67	0.47	2.00	0.40	2.20	0.35
0.35	3.35	0.45	5.80	0.42	1.80	0.47	2.17	0.52
0.35	4.12	0.42	3.47	0.32	2.00	0.40	3.02	0.47

Group VII: (Standard Trauma; Promethazine I. M. 1800 mg over 30-Day Period)

The architectural pattern and cellularity are normal for the first 3 weeks. Very large, frayed fibers are seen during this period, while small fibers are not seen until the second week. A peculiar picture is seen 5 weeks after standard trauma. The epitenon is thickened and cellular. The endotenon is not conspicuous. Tendon bundles are small and contain an increased number of poorly stained nuclei. Most fibers are small and frayed, while some are very large. Many relatively large vessels are seen scattered through the tendon without relationship to the endotenon. The entire tendon stains poorly (Fig. 14). Sections at 8 weeks are similar to those of group II, at 8 weeks. As in group IV, 8 weeks, many small well-stained fibers are seen, associated with large well-stained nuclei and enveloped by cytoplasm. Tendon architecture, fibers, and total number of nuclei seen 16 weeks after trauma are all nearly normal.

Group VIII: (Standard Trauma; Promethazine I.M. 1800 mg over 30-Day Period; Promethazine 15 mg Topically)

For the first 2 weeks after standard trauma tendon architecture is normal, as is the cellularity. Collagenic fibers are generally large and frayed. Small fibers are seen at 1 weeks, the epitenon and endotenon are conspicuous, and

tendon bundles are distinct. There is a paucity of nuclei throughout the entire tendon and the collagenic fibers, which are generally large and frayed, stain poorly. Five weeks after operation, the appearance is similar to that in group II at 2 weeks, and at 8 weeks the tendon resembles that in group IV and group VII, at 8 weeks. The specimens at 16 weeks are similar in all respects to those in group II at 16 weeks.

CONCLUSION

Surgical trauma was recognized to be detrimental to tendon healing as early as 1918 (9), and this fact has been reiterated by many authors. From this study it is evident that minimal surgical trauma to the intact tendon (designated here as "standard trauma") is sufficient to produce an inflammatory reaction and complete loss of normal tendon structure. This finding has made possible the study of the mechanisms and period of time involved in the degeneration and regeneration during healing of such a highly specialized connective tissue as tendon.

As early as 48 hours after standard trauma, there is swelling of collagenic fibers rapidly accompanied by a fraying or dissolving of their margins. Microscopically the fibers have a waxy appearance. The swelling probably represents a rapid uptake of water due to altered pH or electrolyte content of the fibers brought about by enzymes, H-substances, or metabolites released from the injured tissue (10). The frayed appearance of the fiber border probably represents a more rapid loss of interfibrillar matrix than of the fibrila. This may be due to several factors: the hydration and depolymerization of the matrix, the action of hyaluronidase or other metabolites released locally from the injured tenocytes, and, later, from the proliferating cells and the increased blood and tissue fluid in the more cellular and vascular endotenon.

Most fibers originally making up the tendon eventually disappear. This process lasts 5 to 8 weeks.

Cellular changes accompany changes in the fibers. Initially there is a patchy loss of nuclei, followed by an increase in over-all cellularity of the tendon which progresses to a maximum about the fifth week. The cellular proliferation comes from many sources. Cells in the epitenon and endotenon undoubtedly contribute, but many of the new cells arise from cells within the tendon bundles themselves, either from mature tenocytes or from resting primitive mesenchymal cells. One to three weeks after trauma we see increased cellularity in the tendon bundles themselves while the epitenon and endotenon are still relatively acellular. Occasional mitoses are seen within the bundles at this time.

Cells begin to form new collagenic fibers sometime before the eighth week. Such cells have large plumb nuclei and bluish cytoplasm. Intimately associated with the nuclei and partially surrounded by the cytoplasm are newly formed fibers. Initially the fibers are very small, have a smooth border, and stain deeply with eosin.

As the traumatized tendon regenerates, the fibers approach normal size and orientation, while the nuclei of the fibroblasts become smaller. This represents maturation. The epitenon and endotenon return to a resting state, even while cellularity within the bundles is increased. We may speculate that eventually many of these cells will disappear.

As would be expected, vascularity closely parallels the cellular response. Vascularity is first increased in the endotenon as early as 48 hours, and in the epitenon at 1 week. By 5 weeks, the entire tendon is very vascular. Vascularity then declines, but is still increased in the epitenon and endotenon at 16 weeks. Again we may speculate that with time it will return to normal levels.

Thus three phases of reaction to minimal surgical trauma can be recognized, although these phases overlap each other. They may be termed :

1. The degenerative phase, characterized by cell injury and death, with loss of nuclei, and swelling and fraying of collagenic fibers. It may be assumed that during this phase the mucopolysaccharides which serve to cement the collagenic bundles are being depolymerized.

2. The phase of dedifferentiation, characterized by breakdown of the collagenic fibers into the smallest visible fibers, and presumably a depolymerization of the collagen itself into polypeptides and amino acids, thus producing a shift from the formed to the amorphous type of intercellular substance. This is accompanied or followed by a loss of differentiation of surviving tenocytes and a proliferation of young fibroblasts, with increasing vascularity.

3. The phase of growth and maturation, characterized by the differentiation of fibroblasts, formation of new fibers, orientation and increase in size of new fibers, and finally a return to the normal size and structure of the tendon.

Promethazine prolongs the degenerative phase of the response to standard trauma and delays the phase of dedifferentiation. Acceleration of the phase of growth and maturation occurs upon withdrawal of the drug. This effect varies with the route and dosage of the drug administered. Topical promethazine, 300 mg given over 1 day, or 350 mg given over 1 week, have similar effects. Toxic dosages of promethazine (1800 mg given over 1 month), as well as prolonging the degenerative phase, delaying the phase of loss of differentiation, and, upon withdrawal, accelerating the phase of growth hand maturation, alter the time relation of these to one another. This gives rise to bizarre histological findings. A similar bizarre sequence is seen when topical promethazine is added to the regimen.

The following may be a possible explanation of what occurs. In the tendon we find two types of connective tissue : the cellular, less differentiated epitenon and endotenon, and the more differentiated collagenic fibers. The response of the less differentiated connective tissue to injury, whether it be physical or chemical, is the breakdown of cells and intercellular substances, with the release of histamine, serotonin, and other metabolites. These lead to an inflammatory reaction which consists of both proliferative and vascular phases, that is, the formation of granulation tissue. As an end result we have

fibrosis, and possibly overmaturation. Highly specialized connective tissue such as is found in the collagenic fibers follows a similar pattern, except that instead of proliferation there is degeneration. Promethazine as an antihistamine compound may alter the intensity of the inflammatory response either by blocking the breakdown of cells and intercellular substances, by directly inactivating released H-substances, or by competing with them at receptor cells. Thus degeneration of collagenic fibers will be prolonged and there will be little or no stimulation for proliferation of cells. As the effects of the antihistamine wear off, degeneration, dedifferentiation, and maturation proceed at a rapid rate as promethazine itself, in certain concentrations, may act as an injuring agent, hence stimulate the reaction further.

SUMMARY AND CONCLUSION

These studies indicate that minimal surgical trauma applied to an intact flexor tendon produces a marked reaction consisting of a degenerative phase, a phase of dedifferentiation, and a phase of regeneration and maturation.

The histological studies reveal that such highly specialized intercellular substances as collagenic fibers and bundles, though non-living, are not inert and permanent structures. They can break down, completely disappear, and be reformed as a result of the reaction to trauma of specialized tenocytes and undifferentiated mesenchymal cells of the tendon and epitenon.

The effects of an antihistamine (promethazine) are to prolong the phase of degeneration and loss of differentiation and, on withdrawal, to accelerate the phase of growth and maturation. These effects vary with the dose and method of administration of promethazine.

Thus, promethazine has been a useful tool in analyzing the mechanisms of the response of tendon to a standardized trauma as well as indicating a possible approach to the problem of the repair of injured tendons and other highly specialized connective tissue structures.

RÉSUMÉ

Les effets d'antihistamine (Promethasine) à la réaction des tendons à l'égard du traumatisme (étude histologique)

F. G. Walker, S. H. Bensley, W. K. Lindsay

Les expériences faites par les auteurs prouvent qu'un traumatisme chirurgical même le moins important, touchant les tendons des fléchisseurs intacts, déclanche une réaction bien remarquable à quatres phases: celle de la dégénération, de la dédifférentiation, de la régénération et de la maturité.

Les études microscopiques révèlent que les matières à haute spécialization, telles que les fibres et bandes intercellulaires de collagène, malgré leur non-vitalité, ne fonctionnent point en matières inertes et permanentes. Ils peuvent être détruits, ils peuvent disparaître complètement, ils peuvent surgir en tant que résultat de la réaction à l'égard du traumatisme soit des ténocytes spéciaux ou des cellules mésenchymales dédifférenciées des tendons et d'epiténon.

Les effets d'antihistamine (Prométhasine) sont ceux de la prolongation de la phase de dégénération, de la perte de la dédifférentiation et, en même temps, d'accélération

de la phase de régénération et de maturité. Ces effets varient avec la dose et la manière d'application de prométhazine.

Ceci connu, prométhasine devient un aide important dans l'analyse du mécanisme de la réponse du tendon à traumatisme standardisé et, en même temps, elle peut nous aider à résoudre le problème de guérison des tendons injurés si bien que des autres tissus connectives à haute spécialization.

ZUSAMMENFASSUNG

Die Wirkung eines Antihistamins (Promethazine) auf die traumabedingte Reaktion der Sehnen (Eine histologische Untersuchung)

F. G. Walker, S. H. Bensley, W. K. Lindsay

Die vorliegenden Untersuchungen ergaben, dass ein minimales chirurgisches Trauma, an einer intakten Beugersehne gesetzt, eine deutliche Reaktion hervorruft, die in einer degenerativen Phase, einer Dedifferentiationsphase und einer Regenerations- und Reifungsphase besteht.

Die histologische Untersuchung deckt auf, dass so hoch spezialisierte interzelluläre Substanzen wie kollagene Fasern und Bündel, obwohl nicht lebend, dennoch keine inerten und dauernden Strukturen darstellen. Sie können untergehen, vollständig verschwinden und wieder gebildet werden, das alles infolge der Reaktion spezialisierter Tenozyten und undifferenzierter Mesenchymalzellen der Sehne und des Epitenons auf ein Trauma.

Die Wirkung des Antihistamins (Promethazine) besteht in Verlängerung der Degenerationsphase und im Verlust der Differenzierung, und bei Absetzen des Mittels in Beschleunigung der Wachstums- und Reifungsphase. Diese Wirkungen sind von Dosis und Verabreichungsart des Promethazines abhängig.

So erwies sich Promethazine als nützlich für die Analyse des Mechanismus, wie die Sehne auf ein standardisiertes Trauma reagiert; so ergab sich auch ein Hinweis, wie das Problem der Reparation verletzter Sehnen und anderer hoch spezialisierter Bindegewebestrukturen in Angriff genommen werden kann.

RESUMEN

Los efectos de un antihistamínico (Promethazine) sobre la reacción de los tendones al trauma

F. G. Walker, S. H. Bensley, W. K. Lindsay

Estos estudios indican, que el mínimo trauma quirúrgico, aplicado a un tendon flexor intacto, produce una marcada reacción, compuesta de una fase degenerativa, una fase de dediferenciación y una fase de regeneración y maduración.

Los estudios histológicos revelan, que las sustancias intercelulares, tan altamente especializadas, como las fibras colágenas y los manojos de fibras, aunque no vivientes, no son estructuras inertes y permanentes. Ellas pueden romperse, desaparecer completamente, y ser reformadas como resultado de la reacción al trauma de los tendocitos especializados y de las células mesenquimales no diferenciadas del tendon y epitendon.

Los efectos de un antihistamínico (Promethazine) son: prolongar la fase de degeneración y disipar la de diferenciación, y sobre la retirada, acelerar la fase de crecimiento y maduración. Estos efectos varían con la dosis y el método de administración del Promethazine.

De este modo, el Promethazine ha sido un instrumento útil en el análisis del mecanismo de la respuesta del tendon a un trauma estandarizado, así como también una indicación y un posible acceso al problema de la reparación de tendones dañados y de otras estructuras altamente especializadas del tejido conjuntivo.

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Phenergan used in this experiment was kindly supplied by Poulenc Ltd., Montreal. This project was assisted by funds allocated by the Province of Ontario under the National Health Grants Programme of the Department of National Health and Welfare, Ottawa (Dr. Walker and Dr. Lindsay), and by the Banting Research Foundation (Dr. Bensley).

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TREATMENT OF EXTENSIVELY BURNED PATIENTS WITH FREEZE-DRIED HOMOLOGOUS SKIN
(a long-term appraisal of the results)

G. DOGO

The first report on the utilization of 34,485 sq.cm. of freeze-dried skin from cadavers in the local therapy of extensively burned patients was presented to the First International Congress on Research in Burns, held in Washington in September, 1960.

After three years, our experience has extended to 81,615 sq.cm. of skin, employed in the treatment of 25 patients.

The biological material was generously supplied to our Institute by the Tissue Bank Department, U.S. Naval Medical School, Bethesda (Maryland) U.S.A.

I now intend to report on the following points:

the method adopted for the clinical utilization of freeze-dried skin from cadavers;

the histological and clinical results, three years after the healing of some extensively burned patients.

METHOD

The application of freeze-dried tissues on the injured areas is carried out immediately after the elimination of necrotized tissues, on the first appearance of granulations.

The freeze-dried skin is first rehydratated, then mounted on a support of vaseline gauze, and the edges of the single grafts are carefully fitted together or superimposed so as to form one uninterrupted area.

It is important that the sound edges of the lesion should be covered for 1 or 2 cm. with the graft, which are then fixed by a few silk or nylon suture stitches. The injured area can thus be entirely covered. A simple, moderately compression bandaging will fix the graft.

The first dressing is made between the 5th and 7th day. After removing the fragments of dead tissue and cleansing the granulations with simple physiological solution, the procedure of covering with a new lot of freeze-dried

skin is repeated in the manner already described, 24 hours later. That is done 2, 3, 4, or even 8 times.

Another factor we consider to be important is keeping the grafted tissues dry, which is done by accurately covering the granulations, and by previously drying the vaseline gauze and sprinkling the grafts, after application, with a fine layer of penicillin powder or chloramphenicol.

The first application of freeze-dried skin has a cleaning and sterilizing

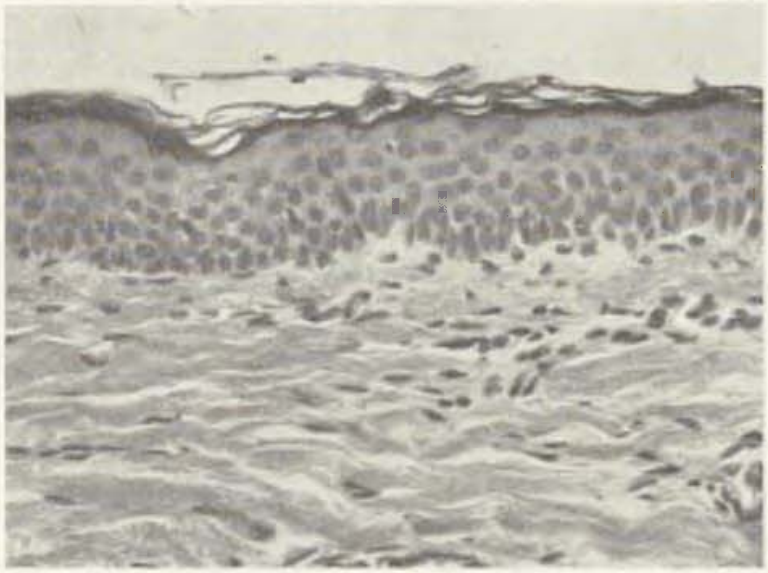

Fig. 1. C.M. (Stain: haematoxylin-eosin) X 350. See explanation in the text.

action on the recipient area; a percentage varying from 30 to 40% of the grafts its destroyed within a week.

The "life" of the grafts successively applied ranges from 12 to 20 days. We have never observed any intolerance reactions.

The benefits the patient receives from these repeated coverings with freeze-dried skin can be listed as follows:

a) decrease or abolition of losses of organic fluids and maintenance of the protein level at values near normal;

b) decrease or disappearance of infectious phenomena;

c) decrease or disappearance of haemorrhagic phenomena;

d) decrease or disappearance of pain;

e) induction of a psychological state of hope and, sometimes, optimism in the patient, sharply in contrast with the dramatic attitudes observed, as a rule, in extensively burned patients;

f) induction of a local healing process which we can describe as "piloted".

In fact, after each application of freeze-dried skin, the extent of the granulation tissue is gradually reduced as the epithelium proceeds from the sound edges towards the centre of the lesion. In certain cases, when — after 4 or 5 successive coverings with freeze-dried skin — the granulations are reduced to

2 or 300 sq.cm., surgical repair with autografts is effected in order to accelerate the healing.

Histological and clinical observations three years after the healing of extensively burned patients

Immediately after the healing obtained by applying freeze-dried skin, the newly-formed tissue appears to be of a rosy colour, not retracted, smooth, but

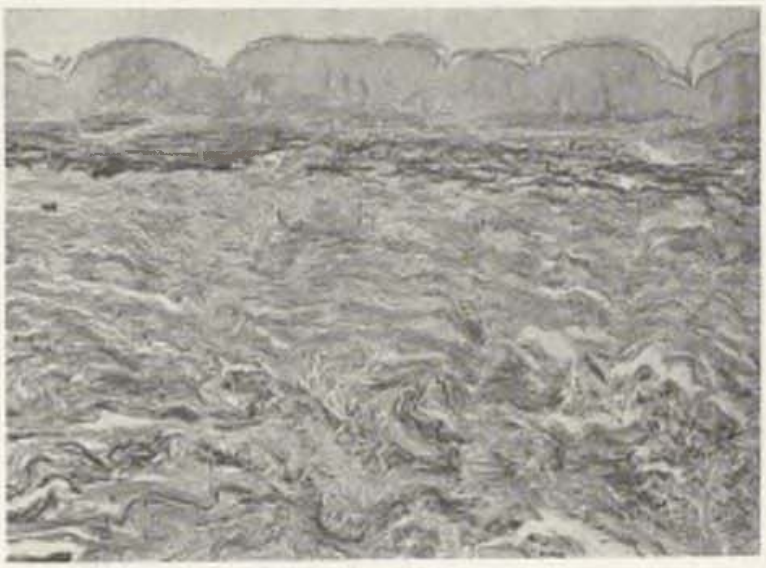

Fig. 2. C.M. (Stain: elastin) X 95. See explanation in the text.

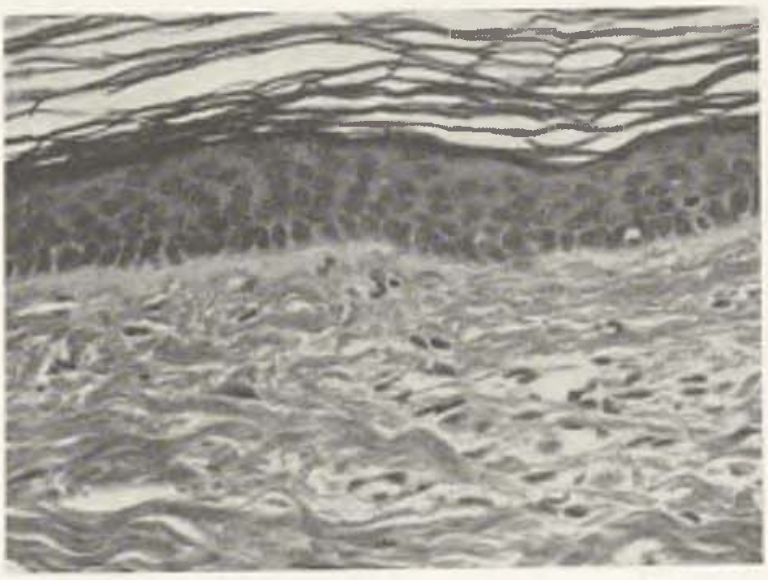

Fig. 3. N.L. (Stain: hematoxylin-eosin) X 350. See explanation in the text.

evenly compact and inelastic. After healing, in the erect posture the lower limbs assume a deep cyanotic coloration.

The picture resembles extensive scar healing, the sclerotic evolution of which might be expected to lead to dramatic problems of reconstruction.

The evolution observed in all the cases treated has, on the contrary, proved to be unbelievably easy and favourable. The compactness of the newly-formed tissues increasingly loosened and moved on the fascial planes. A year later, in all the patients the new skin could be pinched between two fingers and lifted in plicae.

Fig. 4. N.L. (Stain: elastin) X 95. See explanation in the text.

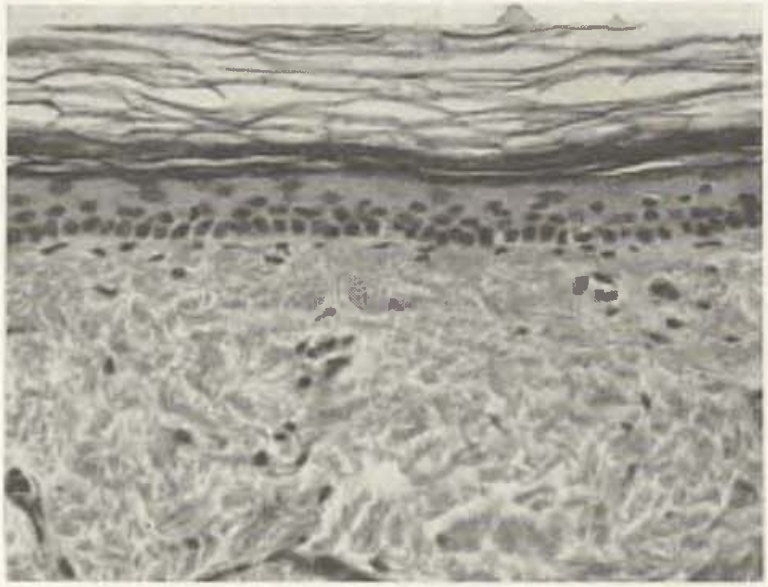

Fig. 5. D.L.P. (Stain: hematoxylin-eosin) X 350. See explanation in the text.

Three years later, the burned areas healed by applying freeze-dried skin appeared to be slightly atrophic, with modest desquamation, but — what is more important — not ulcerated and not retracted. Another common characteristic is a considerable dischromia which clearly separates the newly-formed skin from the normal.

Histological examinations carried out in a number of patients gave similar results. Here are those referring to the three cases illustrated below :

Histological reports

C.M. — Histological examination of a fragment of skin taken from the lumbar region (3 years after healing).

Fig. 6. D.L.P. (Stain: elastin) X 95. See explanation in the text.

The epidermis shows a stratification comparable to normal, with a prickle-cell layer with characteristic morphology ("desmosomes" and easily recognizable fissures among the spinous processes).

The stratum granulosum is limited to one or two rows of cells, however, with little keratohialinic material.

The stratum lucidum consists of a regular eleidinic streak, sinuously following the undulations of the underlying surface.

The stratum corneum consists of a small series of laminae. It tends to be separated from the Malpighian layer and the laminae from each other.

The dermis appears to consist of collagen bundles of a fairly considerable volume, with adequate fibrocytic material. The elastic fibres are numerous near the border with the epidermis, and situated along the larger area of the membrane. More deeply, they lie in different directions and are rather numerous. The tissue is not very vascular; precapillary and venous vessels are however identifiable from the normal structural characters. Lymphoid infiltrations are frequent around the precapillary vessels.

Lastly, there is a scanty growth of the dermal papillae, which is a char-

acteristic of the usual interpenetration between dermal papillae and epidermal gemmae.

In conclusion, we can say that the skin from which the fragment under consideration was taken, is in good condition as far as its entire structural framework is concerned. However, appendages, such as sweat and sebaceous glands, and hair follicles, appear to be absent.

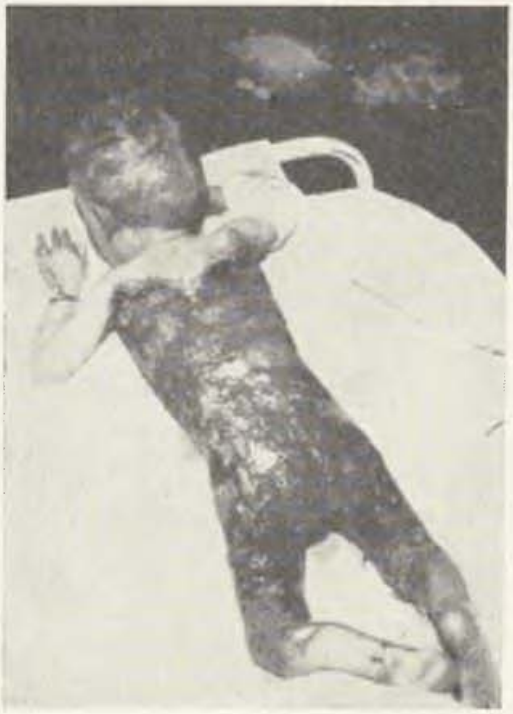

Fig. 7. C.M.

Fig. 8. C.M.

N.L. — Histological examination of a fragment of skin taken from the abdominal region (2 years after healing).

The morphological characters of the epidermis are comparable to those of the skin from the next patient D.L.P., from which they differ only because the thickness is rather greater. The dermis shows a rich collagen framework with rather more considerable vascular material than in the other subjects. Lymphoid infiltrations surround the precapillary vessels.

The elastic material is well represented, though not abundant; near the epidermis it appears to consist of fibres parallel to the surface. There are no skin appendages.

D.L.P. — Histological examination of a fragment of skin taken from the abdominal region (2 years after healing).

The epidermis is very thin in the germinal layer and the elements of the stratum spinosus, which are well identifiable, consist of not more than three or four rows of cells.

The granular layer, lucid layer and corneal layer are identifiable, the latter with a considerable dissociation of the laminae above. There are no dermal

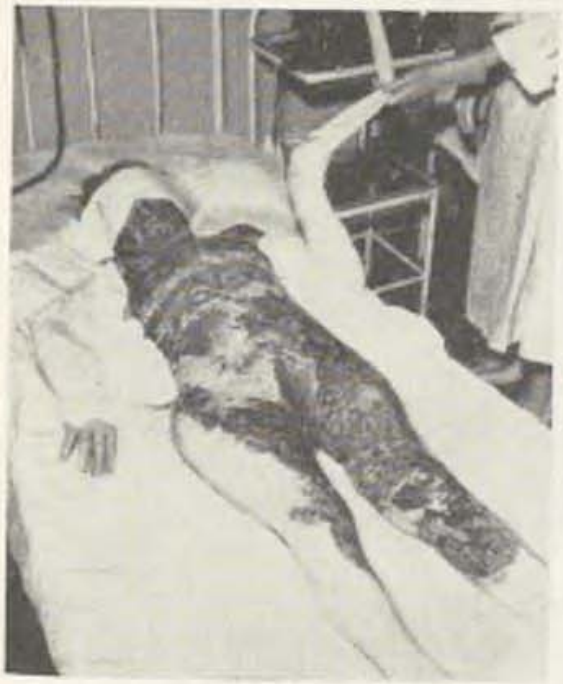

Fig. 9. N.L.

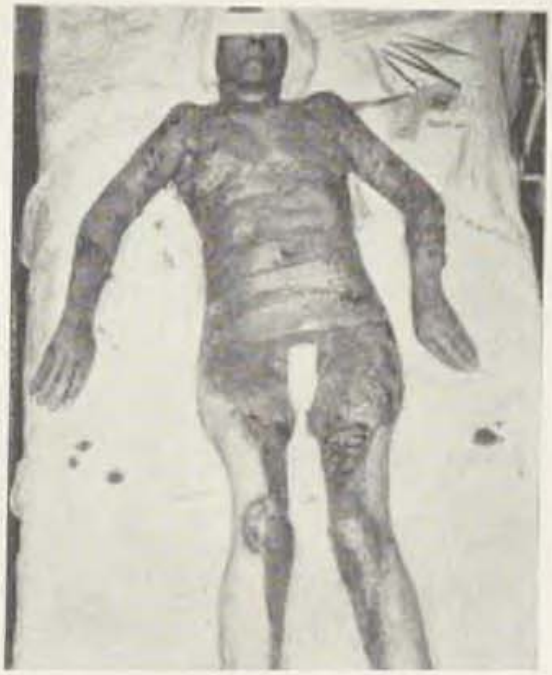

Fig. 10. N.L.

papillae; the structure of the dermis consists of fasciae of small volume, with few fibrocytes and modest capillary material; some lymphoid infiltrations surround the precapillary blood vessels.

An elegant plexus of elastic fibres is present throughout the height of the dermis, with no thickening — unlike the previous cases — near the epidermis.

There are no skin appendages, either glandular or piliferous.

To synthesize, the following fundamental results emerges from the histological reports:

The skin regeneration is brought about by means of an "elementary" skin, having a rather thin epidermis, with a weak cohesion of the planes of the corneal layer, with dermal-epidermal relations deprived of or scarcely provided with papillae and interposed dermal gemmae. Moreover, a remarkable characteristic is the absence — at least up to this stage — of such skin appendages, such as glands and hair follicles.

The vascular material also seems to be rather scarce. It is represented by frequent infiltrations of lymphoid cells, which is probably a reaction to chronic irritating stimuli.

The study of the nervous material is highly interesting, and it will be necessary to devote a further series of investigations to it.

We can assert that a skin has formed having the essential morphological characteristics of a normal fetus between the 6th and the 9th month, without evidence of appendages.

Clinical cases

The results I am now going to report are more significant than any description.

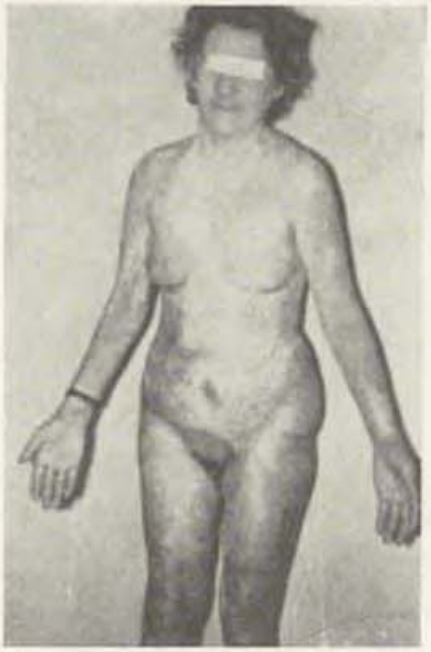

Fig. 11. N.L

Fig. 12. D.L.P.

Case 1, C.M., aged 2. Burns due to boiling water, covering 52% of the body surface. Three weeks after the accident, a granulation area remained covering 40% of the body surface.

Clinically healed in 11 weeks, by utilizing 6,423 sq.cm. of freeze-dried skin in 6 successive applications.

The child, who is in perfect health condition, is now attending the first grade of the primary school.

Case 2, N.L., aged 56. Burns due to fire, covering 65% of the body surface. After five weeks a granulation area remained covering 50% of the body surface.

Clinically healed in 9 weeks, by utilizing 8,760 sq.cm. of freeze-dried skin in 6 successive applications.

The patient has entirely resumed her domestic activities.

Case 3, D.L.P., aged 18. Burns due to fire, covering 60% of the body surface.

After five weeks a granulation area remained covering 55% of the body surface.

Clinically healed in 17 weeks, by utilizing 21,922 sq.cm. of freeze-dried skin in 9 successive applications.

At the time of the accident the patient was employed as a worker in a foundry. Now he is better paid as a business agent and drives his own car.

The patients were discharged from hospital after an average period of two months after complete repair of the injured parts.

None of the patients was submitted to repairing or reconstructive surgery.

CONCLUSIONS

The first question which is spontaneously put when critically considering the above results, is the following: why are not these piloted healings accompained by the so much feared scar sequelae?

Maybe because the exhausted organism is unable to produce connective tissue?

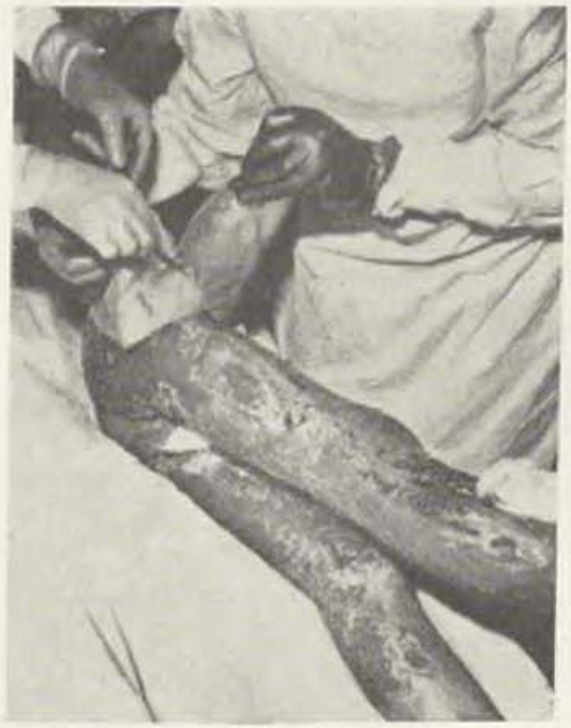

Fig. 13. D.L.P.

Fig. 14. D.L.P.

Or because it is unable to elaborate the necessary elements for its sclerotic evolution?

We do not know.

Or is the lack of scar retraction perhaps due to something present in the freeze-dried (dead) tissues, which are able to depress the production of connective tissue, thus favouring a reconstitution of the skin tissues very near the anatomical and functional norm?

We do not even know this, but it is the object of already planned research.

It has been objected that for each extensively burned patient treated according to the method described here, the quantity of skin required, and therefore its cost, is too great. We can answer back that the number of dead donors is larger than the number of burned patients in need of their skin.

It has also been objected that the average stay in hospital of burned patients treated with freeze-dried skin homografts is longer than the average stay of patients surgically treated with autografts. That might be true — though it has not yet been statistically proved, — but only in the case of patients with burns not exceeding 30%.

The few patients I have referred to you, and all the 25 cases we were lucky enough to able to treat with 81,615 sq.cm. of freeze-dried skin, were extreme cases: with losses of substances exceeding 40%, or even with less extensive burns, but who arrived at our Center in such poor general and local conditions that surgery was contraindicated.

Therefore, we could hardly include these cases in the statistical calculation of the cost and of the stay in hospital of patients with medium-sized burns. In fact, without the aid of freeze-dried skin they would quickly have avoided any statistical investigation, since they would have died.

ACKNOWLEDGMENTS

The author is deeply indebted to Prof. L. Bucciante, Director, Institute of Human Anatomy and Histology, Padova University, for reporting on the histological preparations, and to Dr. M. F. Dolan, Director, Tissue Bank Department, U.S. Naval Medical School, Bethesda, Maryland, USA who very generously supplied him with the freeze-dried material.

SUMMARY

The early covering of the granulations in large burns with biological material is now the most employed method for encouraging the healing of these areas.

The following types of biological material are used:

a) Fresh skin homografts

b) Alternately placed skin homo- and autografts

c) Freeze-dried homologous skin

d) Skin autografts placed on the residual areas of granulations

Experience with the clinical use of 81,615 cm^2 of freeze-dried homologous skin has shown that this material acts as a pilot for epithelium advancing from the edge to the centre of the wound, thereby making more rapid healing possible. The results and experience of using this method in "directed" or "piloted" healing of wounds over a long period are critically evaluated in burns, both in children and adults, and compared with the results obtained on using fresh skin homografts and surgical practice with autografts.

RÉSUMÉ

Traitement de grands brulés avec homéogreffons dermiques lyophilisées. Appréciation des résultats obtenus

G. Dogo

Après des brûlures étendues, la couverture des surfaces de granulation à l'aide d'un matériel biologique et exécutée à temps, représente aujourd'hui la méthode la plus employée pour rendre possible la cicatrisation de ces surfaces.

Types du matériel biologique employé:

a) homéogreffons dermiques frais;

b) homéogreffons dermiques et autogreffons, appliqués alternativement;

c) homéogreffons dermiques lyophilisés;

d) autogreffons dermiques, appliqués sur les surfaces de granulation résiduelles.

Les expériences faites avec l'utilisation clinique de 81 615 cm² d'homéogreffons ont montré que ce matériel agit en tant que pilote de l'épithélium, pénétrant à partir des bords de la lésion vers le centre de la plaie et facilitant ainsi la formation plus rapide d'une couverture.

Les résultats obtenus et l'expérience acquisie par l'utilisation, pendant longtemps, de cette méthode de la cicatrisation «dirigée» ou «pilotée» sur des malades brûlés — enfants et adultes — sont soumis à une analyse critique et comparés aux résultats des cicatrisations, obtenus à l'aide d'homéogreffons frais et à ceux obtenus en chirurgie avec des autogreffons.

ZUSAMMENFASSUNG

Behandlung ausgedehnter Verbrennungen mit lyophilisierten Homohauttransplantaten. Bewertung der Ergebnisse

G. Dogo

Die meistverwendete Methode, das Verheilen ausgedehnter Verbrennungen zu erzielen, ist heutzutage die frühzeitige Deckung der Granulationsflächen mit biologischem Material.

Folgende Arten biologischen Materials gelangen zur Anwendung:

a) Frische Homohauttransplantate;

b) Homohauttransplantate und Autotransplantate in abwechselnder Reihenfolge;

c) lyophilisierte Homohauttransplantate;

d) Autohauttransplantate zur Deckung der restlichen Granulationsfläche.

Unsere Erfahrungen mit der klinischen Verwendung von 81.615 cm² lyophilisierter Homohauttransplantate überzeugten uns, dass dieses Material als Wegweiser für das von den Defekträndern gegen die Wundmitte zu vordringende Epithel dient und so eine schnelle Wunddeckung ermöglicht.

Die Ergebnisse der langjährigen Verwendung dieses Verfahrens einer „gelenkten" oder „pilotierten" Wundheilung bei Patienten mit Verbrennungen — Kindern und Erwachsenen — werden kritisch bewertet und mit den Behandlungsergebnissen verglichen, die mit Hilfe frischer Homotransplantate oder in der chirurgischen Praxis mit Autotransplantaten gewonnen wurden.

RESUMEN

Tratamiento de quemaduras extensas por medio de los homotransplantes cutáneos liofilizados. Valorización de los resultados

G. Dogo

El hecho de que todas las áreas de granulación en casos de quemaduras extensas están cubiertas a tiempo con ayuda de un material biológico, esto representa, actualmente, el método más usado para curar esas áreas.

Clases del material biológico:

a) homotransplantes cutáneos frescos;

b) homotransplantes y autotransplantes cutáneos aplicados alternativamente;

c) homotransplantes cutáneos liofilizados;

d) autotransplantes cutáneos aplicados en las áreas remanentes de granulación.

Las experiencias con la aplicación de los homotransplantes cutáneos liofilizados de 81,615 cm^2 nos han enseñado que este material actúa como un guía del epitelio que avanza desde los bordes del defecto hacia el centro de la herida ofreciéndole de tal manera la posibilidad de cubrir la herida con más rapidez.

Los resultados y las experiencias del uso a largo plazo de este proceder durante la cicatrización „dirigida" o „pilotada" son valoradas críticamente en los quemados — adultos y niños — y comparadas con los resultados de cicatrización logrados con ayuda de los homotransplantes frescos o con los resultados adquiridos a base de la práctica quirúrgica que trabaja con autotransplantes.

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TRANSPLANTATION OF KNEE JOINT
(experimental study)

O. FIALA, V. HEROUT

Homoplastic transplantation aimed at replacing tissues and organs remains an open question. The transplant does not only substitute the damaged organ, but must also take over its function. That is why homologous transplantation of organs is successful in exceptional cases only, e.g. in transplantations between homozygous or heterozygous twins, or in the case of accidental compatibility between the unrelated donor and recipient, or in a recipient with congenital paralysis of immunity (Bücherl 1964). The producing of tolerance in the recipient against foreign material (X-ray irradiation, cytostatics, hormonal therapy, hypophysectomy, etc.) is now the subject of research in homogenous transplantation of tissues and organs. Good results of transplantation in exceptional cases might be a promising factor for the further development of this method of influencing the recipient.

However, bone and cartilage occupy an exceptional position among other tissues. The relatively small density of cells and the presence of inter-cellular substance decrease the antigenicity of these tissues to such an extent that they are not eliminated after transplantation. This provides not only for the possible transplantation of homogenous bone and cartilage tissue itself but also of joint parts or whole joints.

At the beginning of this century already a number of authors tried homogenous transplantation of half or a whole joint both experimentally and in patients (Tuffier 1901, Judet 1908, Lexer 1908, 1909, Axhausen 1919, 1912, Impellomeni 1911, Dalla Vedova 1911, Pucci 1913, Obata 1914). These transplantations were unsuccessful in all cases; break-down of the entire transplant or progressive degenerative changes took place. Even with degenerative changes some functions of the joint could be preserved (Lexer 1909). Later, on the basis of new knowledge and improvement of surgical technique, further attempts at homoplastic transplantation of half or whole joints were undertaken (Payr 1934, Herndon et Chase 1952, 1954, Pap et Krompecher 1956, 1958, 1961, Imamaliev 1958, 1960 a, b, c, Jaroš 1959, Harnach 1959, Kucenok 1959,

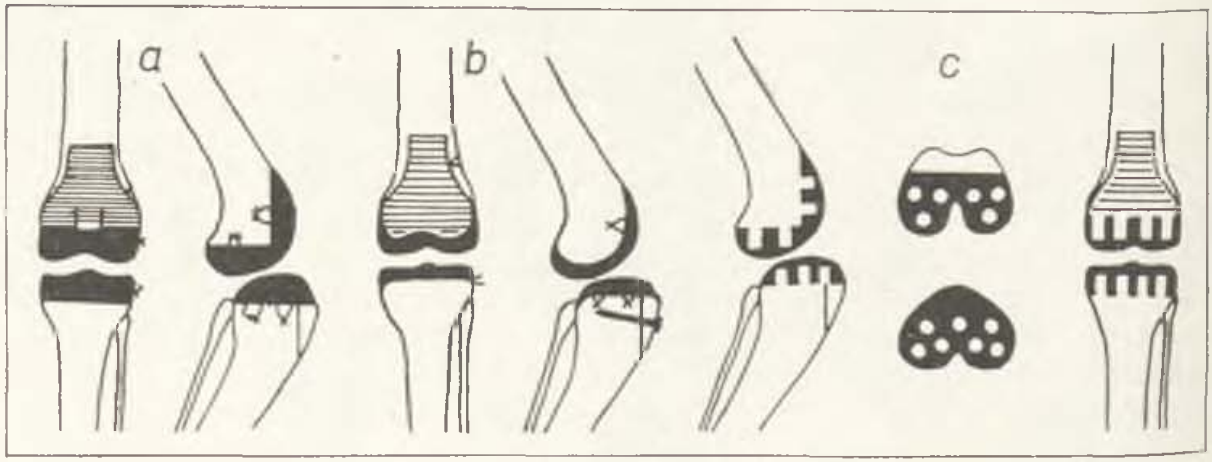
Panova 1960, Erdélyi 1961, 1962 a, b). From the results of these transplantations the following general conclusion can be drawn. Homogenous transplantation of a part of the joint surface or of a not too massive joint end, may lead to quite favourable results; the transplantation of a whole joint, however, is always unsuccessful.

Replacement of the whole joint requires transplantation of three types of tissue: bone, cartilage and connective tissue. The behavour of each tissue following transplantation is specific so that the pre-condition for a successful transplantation rests in creating suitable conditions for preserving the viability or ensuring regeneration of each component. On transplanting homogenous tissue in the form of massive joint, it is, however, necessary to bring the technique of transplantation into harmony with the compatibility of tissues. In this respect no complete success has yet been attained, and that is also why transplantation of whole joints was unsuccessful in most cases and resulted in break-down or a considerable deformation of the joint (Herndon et Chase 1952, 1954, Pap et Krompecher 1961).

That is why in experimental transplantation of whole joints we set ourselves the following four tasks:

1. decreasing immunological response of the organism to the minimum;
2. using exact and perfect surgical technique;
3. ensuring good fixation of the transplant;
4. providing for earliest possible function of the transplanted joint.

We have attempted to decrease the immunological reaction of the organism to the graft by suppressing the antigenicity of the homogenous transplant. For this purpose the graft was stored for a short time, all components rich in cells, such as the articular capsule, the synovial lining, etc. were removed as far as possible, and, of the soft tissues only the ligamentous apparatus was left. Subsequently the bone component of the graft was partly replaced by autogenous bone. The elaboration of an efficient surgical technique and perfect fixation of the graft required many experiments. We tried, in the first



Diagram

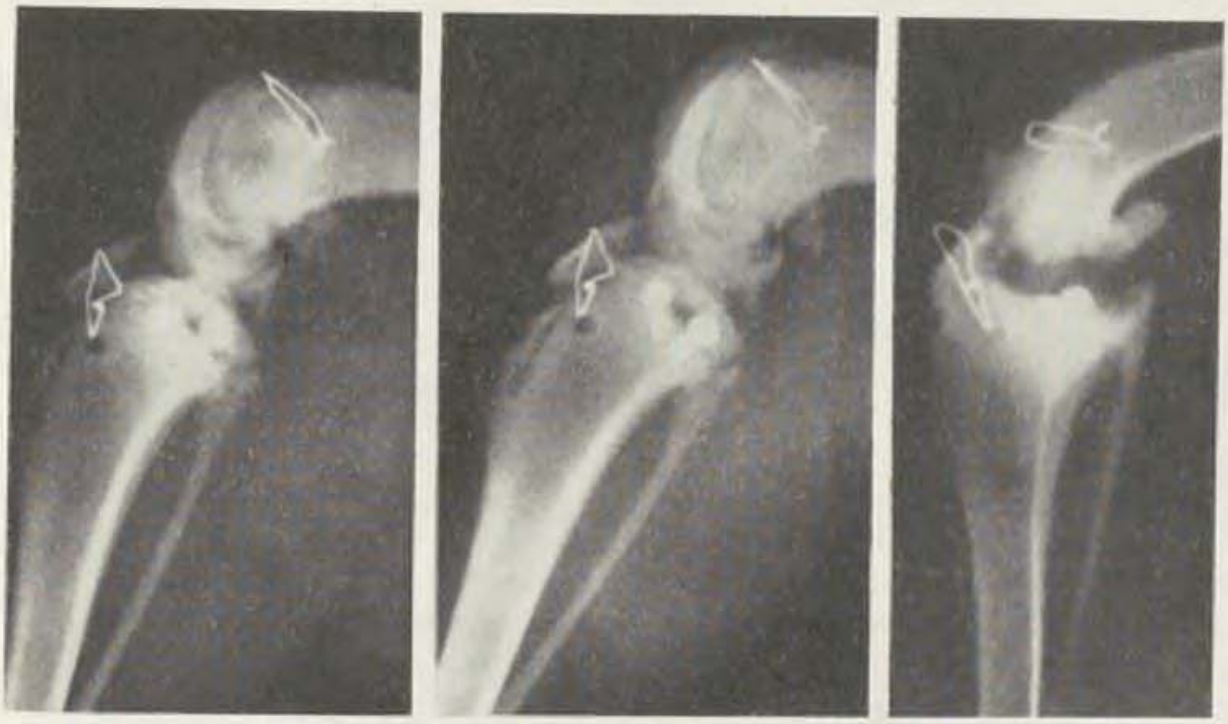


Fig. 1. Dog, No. 224. Lateral X-ray of knee joint after transplantation of homogenous graft with thin layer of subcartilaginous bone. Transplant immediately after transplantation (left picture) does not contact bed exactly; fixed by wire loop and two screws. Two weeks after transplantation, immediately after removal of plaster bandage (centre picture) findings did not differ substantially from those in first picture. Five months after transplantation (right picture) there is distinct disintegration of transplant and considerable deformation of condyles. Screws protrude into joint fissure.

place, methods for safely anchoring the graft and fitting it exactly to the bed, leaving the thinnest possible layer of subcartilaginous bone. This entailed a further task, i.e. to make earliest movement of the operated joint possible.

The first transplantations in 1959 were carried out with the object of finding a suitably large joint and of determining appropriate surgical technique. At the beginning the hip-joint was transplanted, later for facilitating the experiment, the knee joint was chosen, despite its complicated structure.

METHOD

Experiments were carried out in dogs of both sexes and different breeds weighing 16—24 kg. Transplantation of 31 knee joints was carried out. The animal was killed with penthotal and the whole knee joint was removed. At the beginning, we removed the subcartilaginous bone from the graft with a drill so that only a layer 2—3 mm.-thick was left on both parts of the joint graft (Diagr. a). However, this method of processing the graft was abandoned, because the bed for the graft could not be exactly adjusted, even if using a sterile wax cast. That is why we later processed the graft so as to provide straight bone surfaces, even if a thicker layer of subcartilaginous bone had to be left; particularly in the region of both femoral condyles (Diagr. b).

Where the layer of cancellous bone was thickest, bone tissue was subsequently removed with a drill, 4 mm. in diameter, to close under the cartilage and the thus resulting pits filled with columns of fresh autogenous cancellous bone (Diagr. c). Of the soft parts, only the menisci and the collateral and cruciate ligaments were left on the graft. The graft was stored in paraffin oil for 10—14 days, at a temperature of + 4° C. Prior to transplantation, the grafts were washed and left for roughly one hour in a saline solution with added antibiotics. In the experimental animal the knee joint was opened after chiselling off the tibial tubercle, all soft parts (menisci, cruciate ligaments and part of the infrapatellar pad of the fat) removed, and the condyles shaped according to the prepared graft. In the initial experiments only the joint cartilage was removed and the surface of the subcartilaginous bone rounded off with a drill and rasp. In further experiments the joint ends were cutt-off with a saw so as to provide for close contact of the bone surfaces of the graft and bed. The transplant was fixed by wire loops or wire loops plus screws. The lifted tibial tubercle was fixed to the original site by wire loops or loop plus screw. The outer fixation of the knee joint was ensured by a big plaster cast which was left for 10—14 days. After transplantation the animals remained under continuous check-up by X-ray and functional examination from 3 to 26 months. Then the animals were killed and the joints were examined macroscopically; in eight of these, histotopograms of the whole knee joint were made and the individual components, i.e. cartilage, bone and soft parts, were examined histologically.

RESULTS

In the first modification of the experiments, where only the thin osteocartilaginous cover of both joints ends had been transplanted (4 animals), breakdown of the entire graft occurred soon after removal of the fixation. The graft did not have sufficiently close contact with the prepared bed and its thin layer collapsed under the weight of the animal. On opening the joint 3—6 months after transplantation, both parts of the graft were, as a rule, found disintegrated. These parts lay either loose in the articular cavity, or were fixed to the synovial lining or adherent to the joint end so that the shape of the condyles was greatly altered. This deformation was not only due to the break-down of the graft but also to absorption on the surface of the bed, as was particularly obvious around the metal material used. The screws which had been inserted 5—7 mm. under the surface of the processed condyle, slipped into the articular cavity (Fig. 1). Movements in such a joint were very slight.

In one joint, where a thicker layer of subcartilaginous bone had been left, the graft did not break down entirely. The cartilage of the proximal part of the graft healed to the patella and the soft tissues, the wire loop became loose and between the bone of the graft and the processed bone of the femoral condyles contacting each other, there was some movement (Fig. 2). Movements in a thus created joint had a range of 20°.

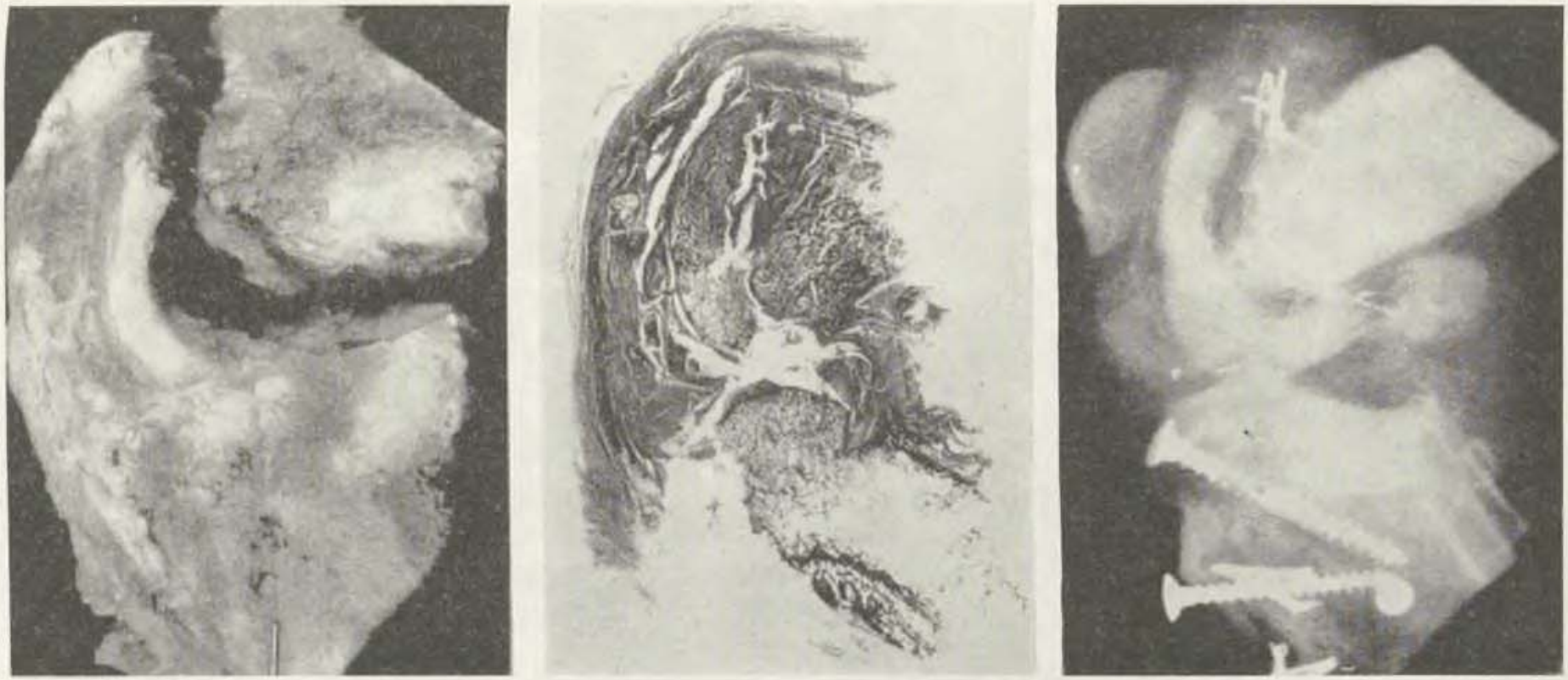


Fig. 2. Dog, No. 261. Knee joint six months after transplantation of imperfectly fixed homogenous graft. Cross-section of joint in sagittal plane (left picture). Proximally preserved section of graft (clear crescent-shaped formation) in front firmly adheres to patella and soft parts. Its posterior bone surface forms a pseudoarthrosis with the processed condyles of the femur. Histotopogram (centre picture), shows distinct unevenness of this false joint fissure. Lateral X-ray (right picture) shows non-disintegrated proximal part of graft and disrupted wire loops (magn: 2×).

In another series of experiments 15 knee joints were transplanted. The graft was adjusted so as to obtain plane bone surfaces that would make possible close contact and firm fixation. Of the 15 dogs only 8 could be evaluated, because in five suppuration set in and 2 died in the kennel from other causes during the experiment.

On observing the operated animals, it was found that the functional findings at different time intervals following transplantation were identical with the X-ray findings. After removal of the plaster the animals limped; later (approximately one month after transplantation) they put weight on the operated limb very well, but at the beginning of the third month they again started limping. Around the sixth month limping became quite obvious and then got worse. The average range of movement (30—60°), however, remained stable.

At the end of the first month after transplantation, the X-ray no longer showed the contact line between graft and bed; the thickness of the graft remained unaltered, the joint surfaces smooth and the joint fissure adequately wide. However, from the third month after transplantation, we could see deformation of the graft in certain places where transformation of the transplanted

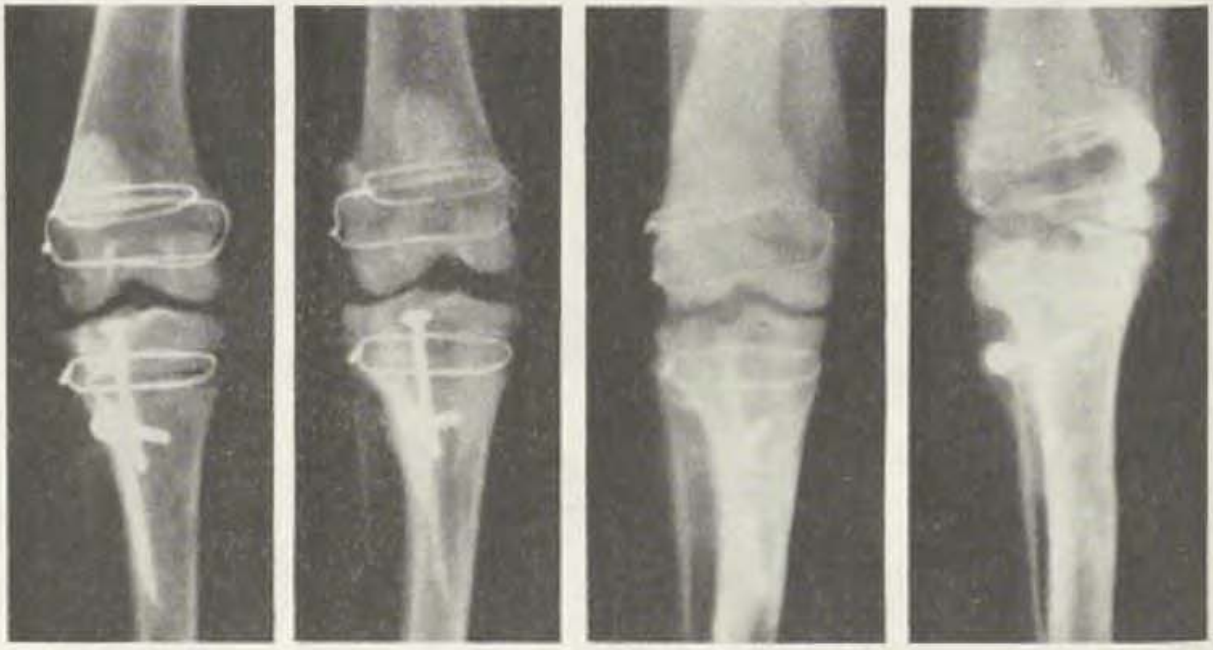


Fig. 3. Dog, No. 245. A-P X-ray after transplantation of massive homogenous knee joint graft. First picture: distinct line of clearing at site of contact between graft and bed (immediately after transplantation). Second picture: joint six weeks after transplantation. Graft has preserved its original shape, line of clearing has disappeared, indicating fusion between graft and bed. Third picture: (six months after transplantation) distinct deformation of lateral femoral condyle, wire loop close under the joint surface, joint fissure substantially narrowed. Fourth picture: joint 15 months after transplantation. Obvious deformation of both femoral condyles and lateral condyle of tibia; wire loops immediately below joint surface. Considerable narrowing of joint fissure and foci of clearing and condensation in condyles.

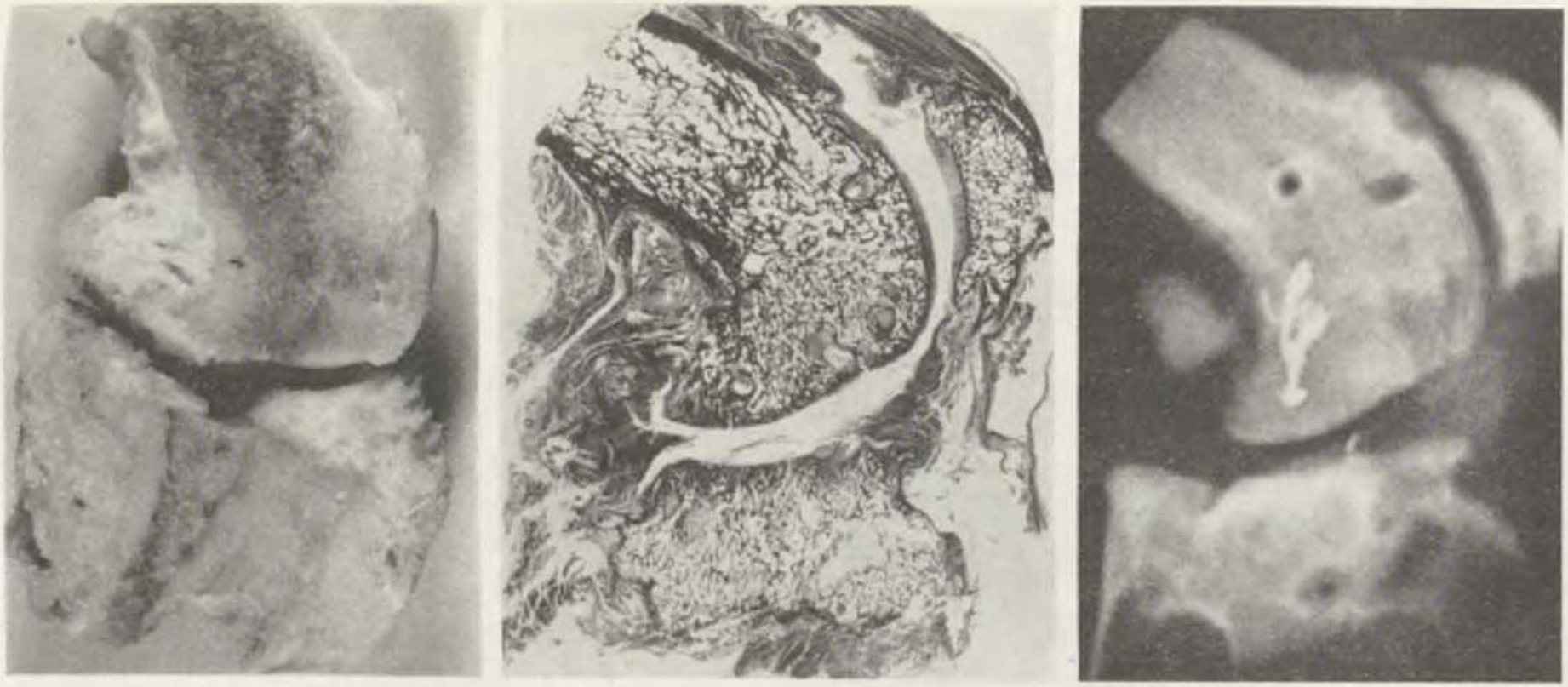


Fig. 4. Dog, No. 252. Knee joint 22 months after transplantation of massive homogenous graft. First picture: cross-section of joint in sagittal plane. Anterior joint surface of femoral part of graft well preserved, covered with cartilage; distal surface, however, abrased and flattened. Histotopogram shows (centre picture) well visible joint fissure with preserved cartilage of both components of patello-femoral joint. Distal surface of femur and joint surface of tibia denuded of cartilage. In marrow of femur several fibrous tissue foci, mainly situated in transplant. Lateral X-ray (right picture) distinctly shows distal parts of femoral condyle flattened by abrasion with progressive deformative changes (magn. 2×).

bone had taken place. This deformation became obvious 6 months after transplantation. Both condyles of the joint graft became thinner, foci of condensation and clearing developed, and in some places there was obvious absorption of the graft. As a result of this, the joint surface became uneven. Most extensive absorption and deformation could be noticed in places exposed to direct pressure and around the foreign material inserted. Changes in the transplant

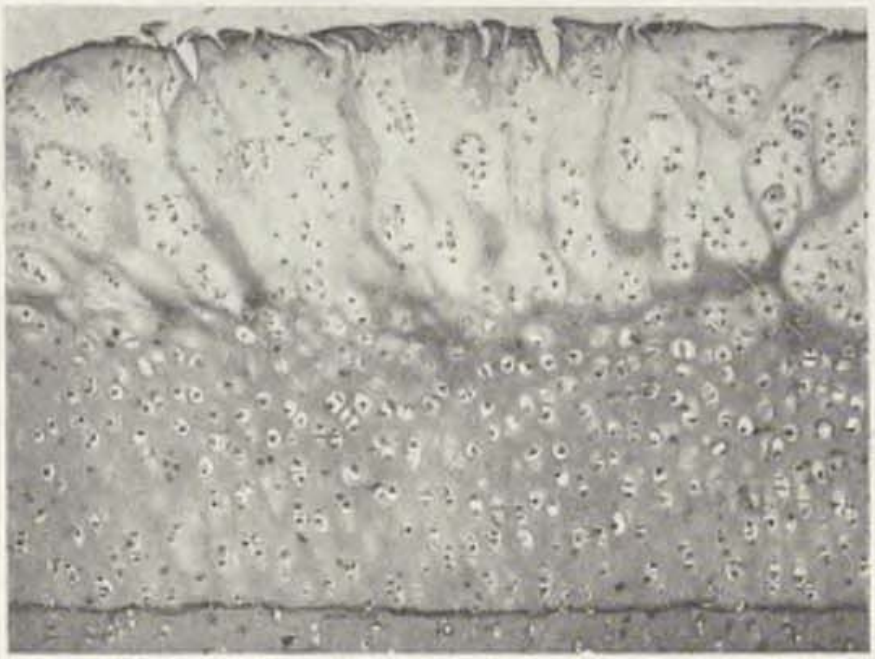


Fig. 5a. Dog, No. 252. Cartilaginous surface of transplant of non-weight-bearing surface of femur. In superficial layer nests of regenerated cartilage cells.

became more pronounced, so that 12 months after transplantation the condyles were considerably altered and serious arthritis deformans developed (Fig. 3). Between the 12th and 24th month after transplantation these changes proceeded very slowly. Such was the finding in all eight animals which could be evaluated, irrespective of whether screws or wire loops had been used for fixing the graft. On opening such a joint 12 months after transplantation, we found a considerably thickened capsule which permitted only a certain range of movements. In the majority of cases the graft was preserved only in places not exposed to direct weight-bearing, i.e. in the anterior aspect of the femoral joint surface. On the condyles of the femur we found an eburnated bone lamella in which fixation material was frequently embedded. In some instances the loops protruded in the articular cavity. On the joint surface of the tibia, there was also eburnated bone, and on the margin considerably narrowed and degenerated menisci. The cruciate ligaments were not preserved. On histological examination we did not find a different structure of bone tissue at the site of fusion, the trabeculae of the bed passed without interruption into the graft. The graft marrow was fatty. The thinned cartilage layer covered only the anterior surface of the femoral part of the transplant, the weight-bearing

part was formed by the abrased bone surface covered with fibrous tissue in which islets of cartilaginous cells could be observed. In the articular capsule fibrous tissue was hypertrophic (Fig. 5a, b).

In two animals, in which the operated knee was stiff, on opening the joint we found quite firm fibrous adhesions between the cartilage of the graft and that of the patella and with the soft parts of the bed. Disruption of these

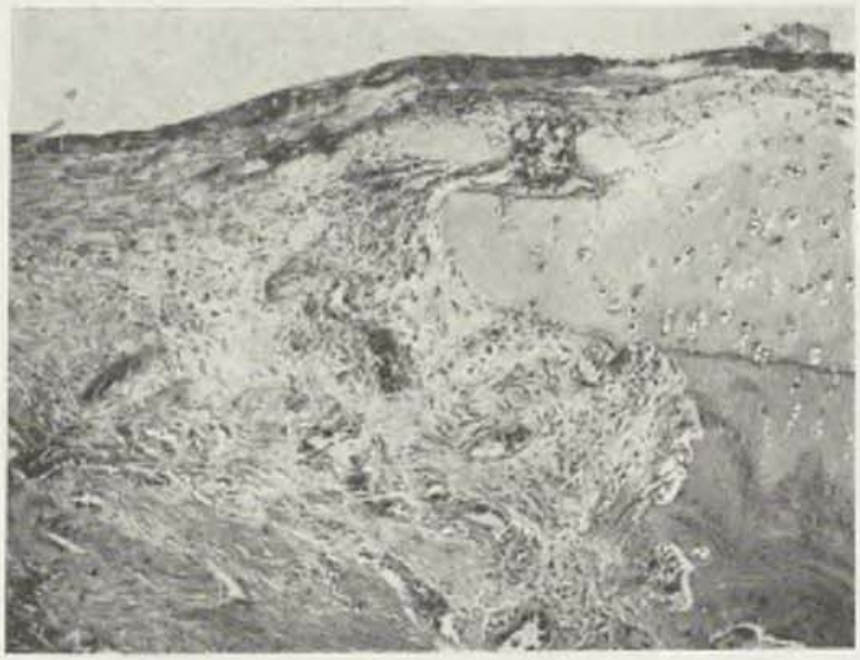


Fig. 5b. Dog, No. 252. Surface of femur at site of weight-bearing and nonweight bearing joint surface. Projection of surface cartilage in state of absorption just into fibrous tissue which covers weight-bearing surface. (Stained with haematoxylin-eosin, magn. 150×).

adhesions was difficult. In such a knee no break-down of the transplant or abrasion of its joint surface had taken place; however, the graft which remained preserved as a whole, had undergone slow absorption. Granulation tissue from the soft parts of the bed grew into the graft at different places and replaced the joint cartilage and the subcartilaginous bone by fibrous tissue (Fig. 6). In the histological picture we found the bony part of such a graft integrated, without any fusion line with the bed. The marrow of the transplant was fatty, the joint cartilage thinner with an uneven surface and necrotic cartilaginous cells (Fig. 7). In some places the cartilage showed ulcerations which reached into the medullary spaces and were filled with connective tissue and fibrous cartilage. The cruciate ligaments were preserved, the cartilage of the menisci showed advanced degenerative changes.

When analyzing the results of transplantation, we realized that it did not suffice only to ensure firm fixation of the graft and its proper contact with the bed, but that it was absolutely necessary to process the thick layer of subcartilaginous cancellous bone in order to accelerate its formation and thus

prevent severe deformative changes. The processing of the subcartilaginous bone, however, should not impair the exact contact of the transplant with and its efficient fixation to the bed.

That is why in subsequent series of experiments we used transplants of the same shape as in the preceding experiments; however, we removed columns of homogenous bone with a bone drill and replaced it with fresh autogenous cancellous bone. Experiments were carried out in 12 animals; nine of these could be evaluated. In six of these nine, part of the transplant broke off as a result of decreased firmness in the graft due to the drill holes made into the bone tissue. In cases where the entire femoral condyle had broken off and had no contact with the bone, gradual absorption took place. The second condyle which had remained in place became considerably deformed as a result of the altered shape of the joint. When the transplant had broken into pieces which became displaced in relation to each other, they fused with the bed, but severe deformation of the condyles resulted.

In these cases later macroscopic and X-ray findings differed from those of the preceding experiments only in their showing more severe deformation of the joint. The newly formed joint surfaces were very uneven, covered in many places by eburnated bone with fibrous adhesions. The broken-off condyles were surrounded by fibrous tissue and formed a component of the considerably thickened capsule. A slight dislocation of the joint thus developed and movements in such a joint became very limited (Fig. 8).

In three animals the graft remained undamaged after transplantation. After the first month, X-ray examination showed disappearance of the line of clearing, indicating the place of contact between graft and bed. Three months after transplantation obvious places of condensation and clearing in the graft could be seen, but no decrease in thickness of its bone component, nor any deformation. Thirteen months after transplantation the graft had preserved its original shape, the joint surface remained smooth, the joint fissure adequately wide. Although the structure of the graft was not entirely uniform, condensation and clearing had mostly disappeared. Twenty-six months after transplantation certain changes were observed in the joint, e.g. arthritis deformans, however, the graft preserved its original shape and the joint fissure remained adequately wide (Fig. 9).

Soon after the plaster had been taken off, the animals started to put weight on the limb, but limped at the beginning; later, however, limping ceased, even on weigth-bearing. One bitch even had puppies.

On opening the joint, we found that the joint cartilage was smooth and glossy. Slight roughness or small ulcerations could be seen, especially on the joint surface which had not been exposed to direct pressure, i.e. on the anterior aspect of the transplant. The menisci and cruciate ligaments were well preserved. The menisci had a slightly yellowish colour. The joint capsule though thickened permitted an almost full range of movements. The fixation material stayed under the joint surface without visible signs of absorption in the surrounding tissue.

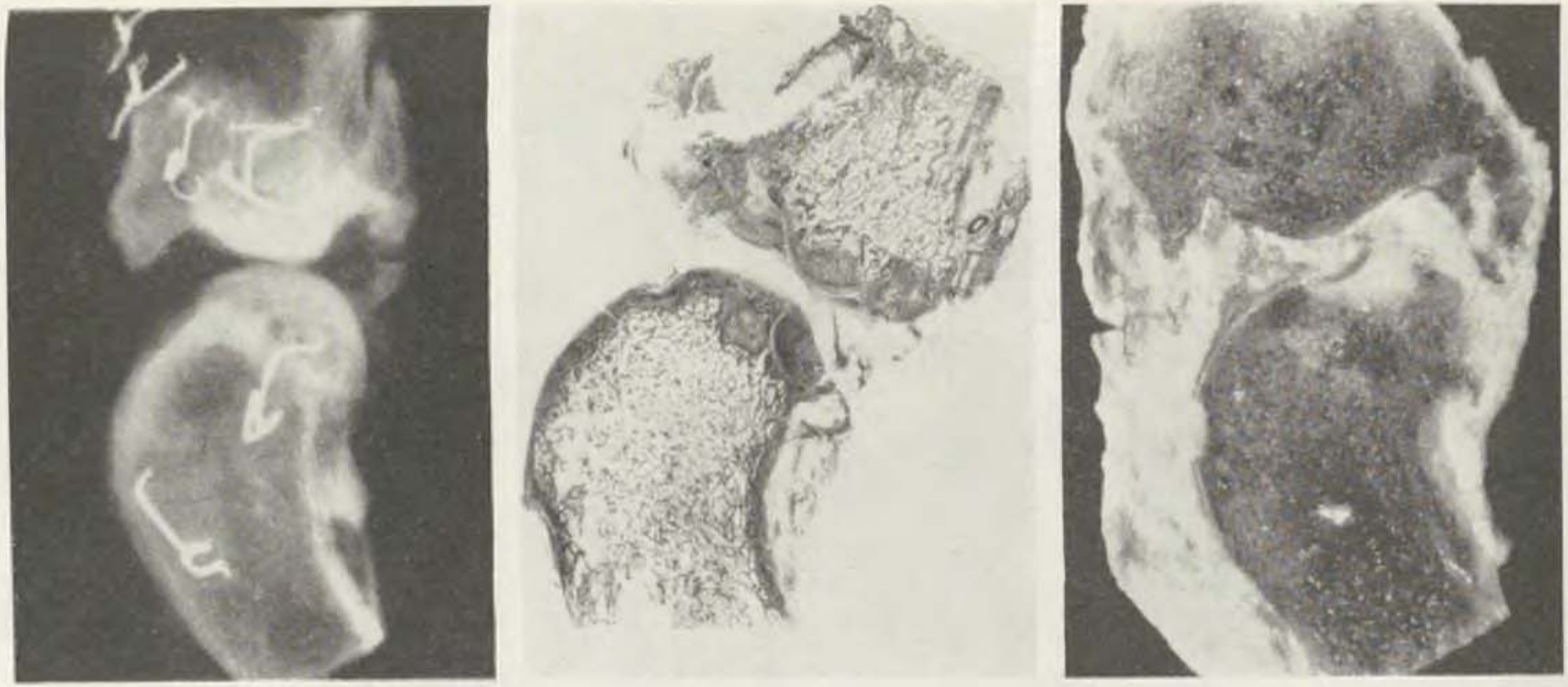


Fig. 6. Dog, No. 335. Fibrous ankylosis of knee joint 10 months after transplantation of massive homogenous graft. On cross-section of joint in sagittal plane (first picture) very good shape of condyles can be seen. Joint fissure, however, is completely filled with fibrous tissue which is adherent to joint cartilage and invades it in some places. Histotopogram (centre picture) after removal of intraarticular fibrous tissue shows defects in the relatively thin cartilage, reaching deep into the subcartilaginous bone. Lateral X-ray (right picture) shows very good appearance of transplanted joint (magn. 2×).

From the histological point of view there was no visible transition from the bed to the bone component of the graft. The cancellous bone of the graft contained mostly fatty marrow and only very few islets of haematopoietic tissue. The articular cartilage of the graft was adequately thick with decreased cell density. The fibrous cartilage of the menisci was well preserved (Fig. 10, 11).

Fig. 7. Dog, No. 335. Picture shows weight-bearing joint cartilage of femur. Cartilage thinner with uneven surface. Most cartilaginous cells necrotis (stained with haematoxylin-eosin, magn. 150×).

DISCUSSION

In our experiments with transplantation of pure homogenous grafts of knee joints we attained the same results as Herndon et Chase (1952, 1954), although we did not use such massive transplants. The best condition of the operated animals was observed 4—8 wekes after transplantation. During that period the animals put weight on the operated joints, did not limp and movement was only slightly limited. On X-ray, the line marking the contact of graft and bed had disappeared. Later progressive deformation of both condyles set in. The most rapid development of these changes could be observed between the 6th and 8th month after transplantation and, as a rule, it continued up to the 12th month. Subsequently the process of deformation slowed down. Joint function began to deteriorate eight weeks after transplantation. The animals started to limp and movements became considerably limited. Only in cases where the joints became stiff, did they not disintegrate.

The results of transplantation of big joint units is thus closely connected with the problem of transformation (absorption-apposition) of the transplanted

Fig. 8. Dog, No. 339. Knee joint six months after transplantation of combined auto-homogenous graft. Cross-section of joint in sagital plane (first picture) shows subluxation due to breaking-off of condyles. Anterior joint surface of femoral part of graft preserved, distal part missing. Joint capsule on posterior aspect is considerably thickened. Histotopogram (centre picture) clearly shows anterior part of femoral transplant which is imperfectly fused with bed, and posterior part shifted backwards. Joint surface of tibia is likewise uneven, preserved only in its posterior part. Its fusion with the bed is also imperfect. Lateral X-ray (right picture) shows torn-off condyles on posterior part of femoral condyle with remnants of broken wire loop. In anterior part of femoral and in remaining part of tibial transplant foci of clearing (magn. 2×).

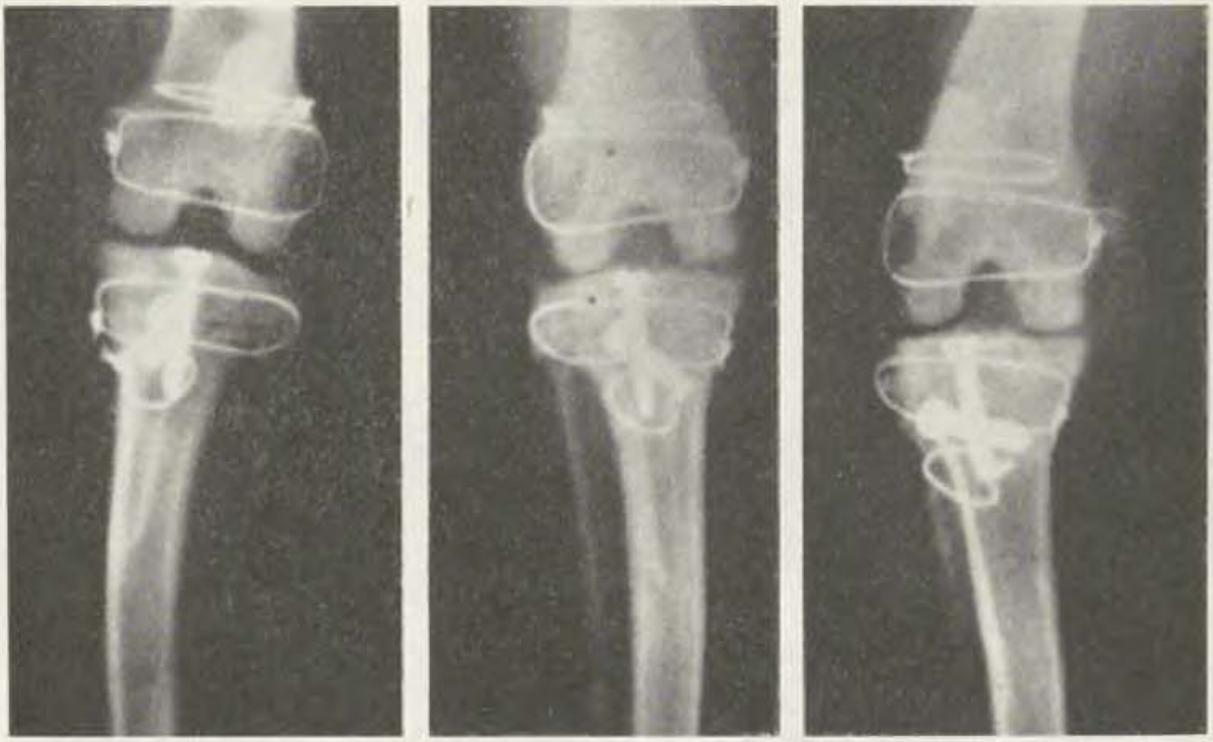


Fig. 9. Dog, No. 177. A-P X-ray of knee joint after transplantation of combined autohomogenous graft. First picture: knee joint immediately after transplantation with distinct line of clearing at site of contact between graft and bed. Second picture (13 months after transplantation): thickness of both parts of graft preserved, joint fissure adequately wide, signs of slight arthritis deformans. Third picture: joint 26 months after transplantation. Original thickness of graft preserved, joint fissure adequately wide. Wire loops remain at same distance from joint surface.

bone (Chase et Herndon 1955). In autotransplantation no substantial deformation occurs, although the entire bone component of the graft undergoes transformation soon after transplantation. Transformation of the graft is rapid, and on the whole, perfect, although a certain change in its shape cannot be excluded. In homogenous transplants a substantial deformation of the graft takes place since its bone component is only slowly and inadequately transformed. The deformation is the bigger the more massive the bone component of the transplant.

That is the reason why recently efforts for improving the results of homotransplantation of joints or their parts have ben developing along two lines. Some authors (Pap et Krompecher 1961) have attempted to preserve the thinnest possible layer of subcartilaginous bone on the osteocartilaginous graft, and, at the same time, to ensure its proper contact to and firm fixation with the bed. This method can only be used in the reconstruction of damaged joint surfaces and in spherical joints (hip and shoulder joints). In replacing a condyle (Imamaliev 1960) or an entire joint (Harnach 1958) it is necessary, however, to use massive grafts. In such a case transplantation can be successful if antigenicity of the graft is decreased in a suitable manner, or, if the

patient is treated so that he tolerates homogenous material. Preparation of the graft by storing, though this partly reduces its antigenicity, does not eliminate the slow and incomplete transformation of the transplanted bone component.

By drilling holes in the cancelleous bone and the homogenous graft and filling them with autogenous cancellous bone, we have attempted to reduce both the volume of the homogenous tissue and, by producing a larger surface and introducing autogenous tissue, to speed up and improve the process of absorption and apposition in homogenous transplants, as well as to improve the unfavourable shape of the graft. Pap et Krompecher (1961) have pointed out that the shape of the graft might also influence the result of transplantation. It is of advantage if the transplant is flat and the size of its cartilaginous surface corresponds to that of the surface of the bone component, which makes contact with the bed. If, however, the joint surface of the transplant is convex in two planes (e.g. femoral condyles) the surface of the bone contacting the bed is then too small for adequate revascularization with regard to the volume and the surface of the graft.

In our experiments the demand for a large contact surface in grafts of the distal femoral condyles, where the angulated shape created a relatively large surface for revascularization and transformation of the bone component,

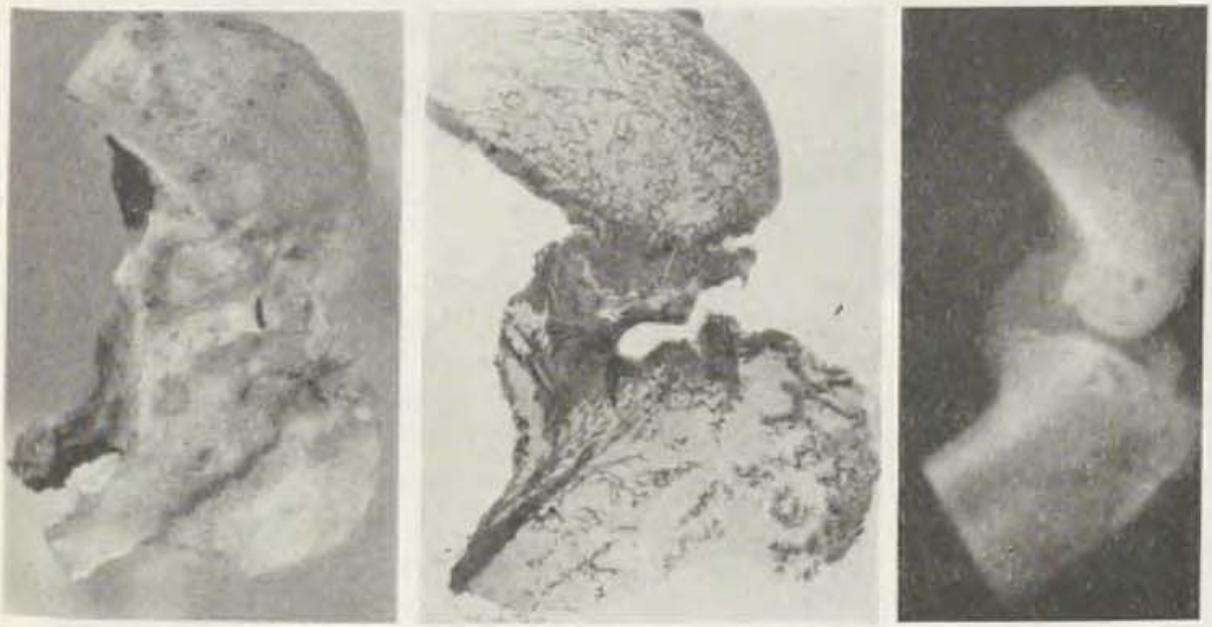


Fig. 10. Dog, No. 177. Knee joint 26 months after transplantation of combined autohomogenous graft. Cross-section of joint in sagittal plane (first picture) shows good shape of condyles. Femoral joint surface covered by well preserved cartilage. Both parts of transplant linked by curciate ligaments (anterior part and thickening of posterior capsule). Histotopogram (centre picture) shows bone component of the transplant of same structure as bed, transition line not visible. Joint cartilage on femoral condyle adequately thick; on tibia cartilage replaced by fibrous tissue. Lateral X-ray (right picture) clearly shows good shape of condyles, particularly the smooth surface of femoral condyles (magn. of histotopogram 2×).

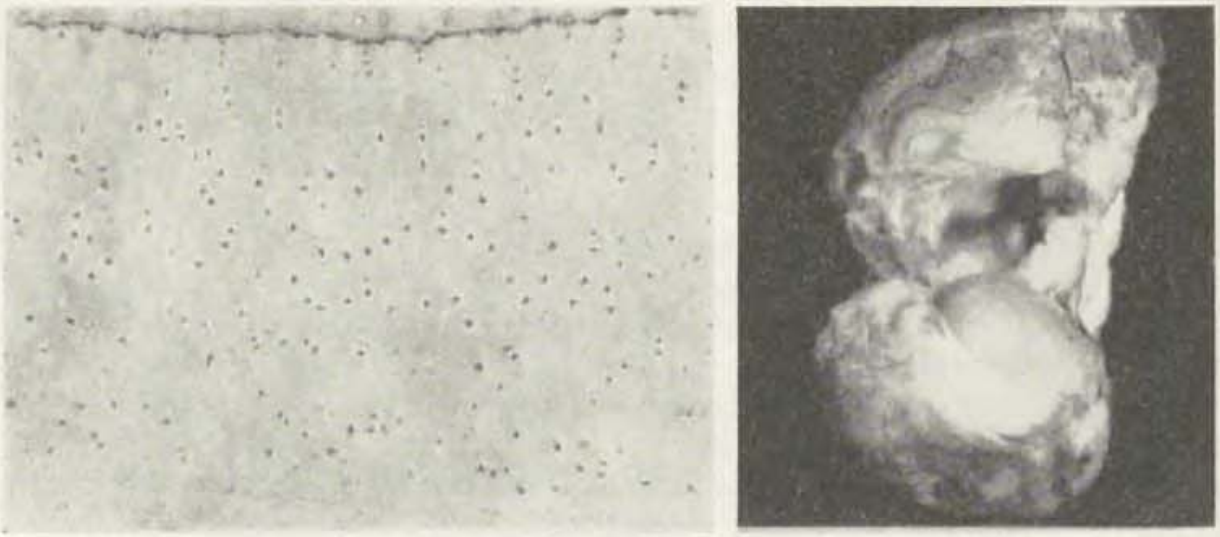


Fig. 11. Dog, No. 177. Macroscopic view of open joint (left picture). At top condyle covered with smooth cartilage; below well preserved meniscus and cruciate ligament. Right picture shows weight-bearing joint cartilage from femoral part of transplant. Cartilage adequately thick, basal layers adequately rich in cells. On surface, shadows of necrotic cartilaginous cells (stained with haematoxylin-eosin; magn. 150×).

was met. However, since the tibial part of the transplant was cut off at right angles to the long axis of the bone, which left a thicker layer of subcartilaginous bone, the contact surface of the transplant was smaller than the joint surface.

It might be assumed that the angulated shape would have been also more suitable for that part of the graft.

Removal of homogenous bone by drilling creates a further problem. We decreased the firmness of the graft which led to damage on weight-bearing. We were not surprised to find disintegration where a greater number of holes had been drilled; however, it was interesting to find that in some cases even a graft with few burr holes had broken down, whereas in others it had remained intact. We found that the mechanical firmness of the graft partly depends on the slant of the bed. If the anterior surface of the femur is cut off obliquely, the mechanical force acts directly on the site of least resistance, i.e. where two bone surfaces meet at right angles. That is why abruption of one or both condyles takes place on weight-bearing. If the anterior joint surface is cut off parallel with the longitudinal axis of the femur, the bone surface of the condyles contacts this axis almost at right angles and thus mechanical damage of the graft on weight-bearing occurs less frequently. On observing our three animals in which a combined graft (a homogenous graft with columns of autogenous bone) had been employed with success, we found a considerable similarity in X-ray and macroscopical findings with the results after retransplantation of a knee joint (autogenous graft) carried out by Herndon et Chase (1952). Although the X-ray, taken eight weeks after transplantation, showed distinct foci of clearing and condensation, the thickness of the graft remained the same and the joint fissure adequately wide. After 2 years we found slight deformative

changes. On opening the joint we found only few ulcerations covered with pannus on the anterior surface of the transplanted cartilage, and on the condyles slightly roughened cartilage in some places; the menisci were macroscopically well preserved and the cruciate ligaments glossy and firm. Ulceration of the joint surface was found in places which did not bear any weight, i.e. on the anterior aspect of the joint surface of the femur, opposite the border of the patella. Similar findings were reported by Herndon et Chase (1954) and Pap et Krompecher (1961). The latter authors ascribe considerable importance to the function of the joint after transplantation of the joint surface. They maintain that the actual reason for the failure of homotransplantation — tissue incompatibility — cannot be determined merely by the way in which the recipient tolerates the transplanted tissue at any site in the organism (bone and cartilage grafts in the subcutaneous tissue, the muscles or the peritoneal cavity). The authors believe that the transplants do not react properly unless functionally loaded after transplantation. This means that not even a transplant is an exception of the fundamental biological rule, i.e. that the organism only preserves a functioning organ; an organ that is not active is absorbed or eliminated.

In our experiments stiffness occurred in two operated joints after homogenous transplantation. We assumed that the appearance of such a joint would be bad. It is interesting to note, however, that no disintegration of the transplant occurred in these cases even after 12 months. Tough fibrous tissue surrounded the transplant and was also firmly adherent to its cartilage, in some places it gave rise to deep ulcerations of the joint surface. The joint was obviously saved from direct weight-bearing and that is why it did not disintegrate on transformation of the bone component. The joint cartilage which did not perform its function had become necrotic, in many places absorbed and replaced by fibrous tissue, as happens in transplantation into soft tissue.

However, it is very difficult to produce correct functional loading of the transplanted joint in experiments. If the fixation bandage is removed, the animal does not only start moving the limb but also puts weight on it. Since during transformation of the graft the cartilage has no adequate support of subcartilaginous bone, it disintegrates in places where weight-bearing is excessive. This results in damage of a large section or the entire graft and thus deformation of the joint. By a combined graft we wanted not only to decrease the antigenicity of the stored homogenous bone, but also to eliminate the possibility of graft deformation by early functional loadening. We assumed that the columns of autogenous bone would be transformed before firmness of the graft is endangered by the transformation of the homogenous bone (approximately 2—3 months after transplantation).

Burwell (1964) tried to create a homogenous graft of properties inherent in autotransplants by injecting autogenous red marrow into homogenous cancellous bone from which the marrow had been washed out. After transplantation of such a combined graft into the soft tissue in mice there was no im-

munological response of the host; moreover stimulation to produce and transform bone was much greater than in the bone itself.

We confirmed the decisive importance of weight-bearing for preservation or destruction of the transplant in experiments with both pure homogenous and combined auto-homogenous grafts. In each experiment, where dogs heavier than 18 kg. were used, damage to the graft occurred or it disintegrated entirely. Not only did the graft break, but often also the metal wire by which the transplant was fixed to the bed. In smaller dogs, the wire loops were not damaged, providing that the graft did not disintegrate as a result of bad preparation. Three successful experiments with the use of auto-homogenous grafts were carried out in animals weighing 16—18 kg.

The use of homogenous osteocartilaginous grafts in reconstruction of entire joints or massive condyles requires further study. It will be necessary, first of all, to bring transformation of the homogenous bone component of the graft closer to that of autogenous bone. One possibility is the employment of combined auto- and homogenous grafts. In such a case it will be necessary, however, to provide technical conditions which would prevent this combination from impairing the firmness of the graft as a whole; this would be possible either in the form of an absolutely perfect substitute for the drilled off homogenous bone component by autogenous cancellous bone or by impregnating the medullary spaces of the homotransplant with autogenous marrow, as reported by Burwell. Another alternative is to develop transplantation tolerance for the duration of the transformation of the graft. Both possibilities will be the subject of further experiments.

CONCLUSION

On the basis of the literature and our own experience, the following principles can be laid down for the transplantation of entire joints in the form of homogenous osteocartilaginous grafts:

1. To ensure the exact fitting of the graft to the bed even if a thicker layer of subcartilaginous bone has to be left; any small inexactitude leads to worsening of results.

2. To make the area of the contact surface of the graft approximately the same as that of the surface of the transplanted joint cartilage. With a small contact surface between graft and bed conditions for vascularization and transformation of the graft are greatly impeded.

3. To fit the graft to the bed so that there will be the smallest possible mechanical strain to the least resistant parts of the graft after discontinuation of immobilization. If this is not adhered to, it leads to abruption of part or disruption of the entire transplant.

4. To fix the graft absolutely firmly to the bed. On imperfect fixation, movement and weight-bearing of the transplanted graft leads to its disintegration.

5. To provide for early functional loadening of the operated joint. This is possible by observing the afore-mentioned conditions.

6. To reduce the immunological reaction evoked by foreign tissue and thus to bring the transformation of the bone component of the homogenous graft closer to that of autogenous tissue. This can be attained either by treating the graft or the recipient. Efficient preparation of the graft might be ensured by a suitable combination of homogenous and autogenous tissues, and that of the host, by producing transplantation tolerance for the duration of the transformation of the graft. Imperfect transformation of the graft is the main reason for its deformation and disintegration.

7. To preserve the viability of the transplanted cartilaginous component by very sparing storage, by not exceeding expiration time, and by creating suitable conditions for its survival after transplantation. Failure to observe these conditions leads to disintegration of the transplanted joint cartilage and thus to disintegration of the bone component of the graft.

SUMMARY

In 31 dogs entire knee joints were transplanted in the form of stored homogenous osteocartilaginous grafts. In 19 of these animals the knee joints were transplanted as a pure homogenous graft, in 12 as a combined graft. In these, columns of homogenous bone were replaced by fresh autogenous cancellous bone. The animals remained under continuous X-ray check-up and functional examination for 26 months after transplantation. In eight dogs the findings were evaluated microscopically.

In four animals, where a pure homogenous graft with only a thin layer of subcartilaginous bone had been transplanted, the graft disintegrated soon after discontinuation of immobilization. The reason for this early break down was a poor fit between the graft and its bed. In 15 dogs transplantation of massive homogenous grafts with broad contact surfaces with the bed was carried out. In most of these dogs disintegration of the graft and considerable deformation of the condyles took place, due to imperfect transformation of the subcartilaginous bone of the graft. Only in two dogs of this group, which developed fibrous ankylosis, was the original shape of the bone component of the graft preserved; however, the articular cartilage underwent necrosis and was gradually absorbed.

Transplantation as successful in three out of the 12 animals in which a massive auto-homogenous graft was transplanted. In these three, movements of the operated joint two years after transplantation were only slightly limited; the animals were able to put full weight on the limb. X-ray examination showed signs of a slowly developing arthritis deformans. The cartilage of these joints was mostly smooth and glossy, cell density, however, was markedly decreased when compared with normal cartilage. The cruciate ligaments and menisci were well preserved.

Principles for transplantation of entire homogenous joints are laid down.

RÉSUMÉ

La transplantation des articulations du genou (Travail expérimental)

O. Fiala, V. Herout

La transplantation des articulations du genou en forme des transplants ostéo-cartillagineux homogènes conservés a été pratiquée à la grouppe de trente-uns chiens. De ceux-là, dix-huit chiens ont reçu l'articulation du genou sous forme du transplant homogène pure, tandis qu'à douze chiens on a transplanté des transplants composés. Dans ce cas, les colonnes de la partie osseuze homogène ont été remplacées par le frais tissu spongieux autogène. Les animaux ont été examinés quand à la fonction si bien que par les rayons X durant les vingt-six mois suivant l'intervention. Les données microscopiques ont été pratiquées chez huit des chiens.

Les quatres animaux ayant reçu le transplant homogène pure dont la couche osseuze sous-cartillagineuse était mince, ont marqué la décomposition du transplant sitôt l'immobilisation terminée. La cause en était l'impossibilité respective de l'assimilation du transplant à l'égard de sa couche. Un transplant massif homogène doué d'un grand plan communicatif à l'égard de sa couche fut appliqué aux quinze des chiens. La plupart d'eux a marqué une forte décomposition du transplant et mutilation de la partie articulaire grâce à la transformation inadequate de l'os sous-cartillagineux du transplant. Ce n'est que chez deux chiens de cette groupe, ayant formé une ankylose fibreuse de l'articulation, que la partie osseuze du transplant a conservé l'aspect original, le cartillage articulaire au contraire nécrotisé petit à petit fut, à son tour, victime de la résorption.

Les douzes animaux ayant reçu un transplant massif auto-homogène, ont marqué une transplantation réussie dans trois des cas. Deux ans passés, les mouvements de l'articulation opérée ont été limités très légérement bien que les animaux s'en étaient servi a leur gré. Les rayons X n'ont découvert qu'une légère arthrose déformative en stade de développement. Le cartillage articulaire respectif était de la plupart lisse, brillant, mais sa richesse en cellules, a l'égard de celle du cartillage normal, était moins exprimée. Les ligaments croisés et les ménisques articulaires étaient en état excellent.

Les règles de la transplantation de l'articulation homogène vient d'être donnés.

ZUSAMMENFASSUNG

Transplantation des Kniegelenks (Experimentelle Studie)

O. Fiala, V. Herout

Bei 31 Hunden wurden ganze Kniegelenke in Form konservierter homogener Knochen-Knorpel-Transplantate übertragen. Dabei wurde bei 19 Tieren das Kniegelenk als rein homogenes Transplantat, bei 12 Tieren als kombiniertes Transplantat übertragen. In den letzteren Fällen wurden abgetragene Streifen des homogenen Knochens durch frische autogene Spongiosa ersetzt. Die Tiere wurden funktionell und röntgenologisch bis zu 26 Monaten nach der Transplantation verfolgt. Bei acht Hunden wurden dann die Ergebnisse mikroskopisch untersucht.

Bei 4 Tieren, bei denen ein rein homogenes Transplantat mit einer nur dünnen subchondralen Knochenschicht übertragen wurde, kam es bald nach Entfernung der Immobilisierung zum Zerfall des Transplantats. Der Grund für den frühzeitigen Zerfall beruhte in dem Umstand, dass das Transplantat nicht vollkommen an den Knochenstumpf adaptiert werden konnte. Bei 15 Hunden wurde ein massives homogenes Trans-

plantat mit grosser Berührungsfläche gegen den Knochenstumpf übertragen. Bei der Mehrzahl dieser Tiere kam es zum Zerfall des Transplantats und zu bedeutender Deformation des Gelenkendes auf Grund eines unvollkommenen Umbaus des subchondralen Knochens des Transplantats. Nur bei zwei Tieren dieser Gruppe, bei denen sich eine fibröse Gelenksankylose bildete, behielt der transplantierte Knochen seine ursprüngliche Gestalt, der Gelenksknorpel wurde jedoch nekrotisch und resorbierte sich allmählich.

Bei 12 Tieren, denen ein massives Auto-Homotransplantat übertragen wurde, war die Übertragung in drei Fällen erfolgreich. Bei diesen Tieren war zwei Jahre nach der Transplantation die Beweglichkeit des operierten Gelenks nur wenig eingeschränkt, die Tiere konnten die Extremität voll belasten. Röntgenologisch wurden Zeichen einer sich langsam entwickelnden deformierenden Arthrose festgestellt. Der Gelenksknorpel eines solchen Gelenks war zum grössten Teil glatt, glänzend, sein Zellgehalt war jedoch im Vergleich mit normalen Knorpel bedeutend herabgesetzt. Die gekreuzten Bänder und Menisken waren gut erhalten.

Es wurden Grundsätze für die Übertragung ganzer homogener Gelenke aufgestellt.

RESUMEN

La transplantación de las articulaciones de la rodilla (Estudio experimental)

O. Fiala, V. Herout

En 31 perros fueron transplantadas todas las articulaciones de la rodilla, en forma de injertos osteo-cartilaginosos homogéneos conservados. De esto, en 19 animales, la articulación de la rodilla fué transplantada como un injerto homogéneo puro, en 12, el injerto fué combinado. En estos casos, las columnas taladradas de la porción homogénea del hueso, fueron reemplazadas por hueso espongioso fresco y homogéneo. Los animales fueron observados funcionalmente y por medio de la radiografía, hasta los 26 meses después de la transplantación. En 8 perros, los hallazgos fueron después valorizados microscópicamente.

En 4 animales, donde fué transplantado un injerto homogéneo puro solamente con una capa delgada de hueso subcondrial, se produjo la desintegración del injerto, pronto después de la eliminación de la inmovilización. El motivo de la desintegración temprana, se debió a la imposibilidad de una adaptación completa del injerto con respecto a su lecho. A 15 perros se les transplantó un injerto homogéneo masivo con una gran superficie de contacto con respecto al lecho. En la mayoría de estos animales, esto condujo a la desintegración del injerto y a una considerable deformación del extremo articulador, teniendo como fundamento una restauración incompleta del hueso subcondrial del injerto. Solamente en 2 animales de este grupo, donde se formó una anquilosis fibrosa de la articulación, la porción ósea del injerto conservó su figura original, sin embargo murió el cartílago de la articulación y fué gradualmente reabsorbido.

En 12 animales, en los cuales fué transplantado un autohomoinjerto masivo, la transplantación fué exitosa en tres casos. En estos animales, el movimiento de la articulación operada, 2 años después de la transplantación, fué solamente un poco limitado, los animales pudieron utilizar completamente su extremidad ante cualquier carga. Por medio de la radiografía, fueron observados lentamente, los signos de una artrosis deformativa. El cartílago de tal articulación, en su mayor parte, apareció liso, brillante, sin embargo su celulización fué esencialmente más baja en comparación con el cartílago normal. Los ligamentos cruzados y los meniscos se mantuvieron bien conservados.

Están determinadas las bases para la transplantación de toda la articulación homogénea.

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THE TASKS OF THE TISSUE BANK IN CLINICAL TRANSPLANTATIONS

R. KLEN

I shall report on the tasks of the Tissue Bank in clinical transplantations as I see them after more than 12 years of work in our Tissue Bank, although the range of this Tissue Bank is much greater (1).

The Tissue Bank is primarily a laboratory which undertakes the routine securing, preparation, examination, preservation and distribution of all tissues used in clinical transplantation; secondly, it is a research centre; thirdly, on the basis of both above functions, it provides lectures and courses and, fourthly, it can be charged with same organization tasks (2).

The Tissue Bank is mainly engaged in routine work which finds a wide field of application in reconstructive surgery. The most frequent application is found by grafts of compact and/or spongious bones, fascia, dura mater, cartilage, skin and cornea. Less frequently, kidney, transplants of joints, parts of joints, and amnion-, tendon-, and vitreous body- transplants are used. Thus far, transplants of bone marrow, endocrine glands, tooth germ etc. have only been used in few cases. Vascular homotransplantation, which was often performed a few years ago, has been completely replaced by alloplastic prostheses. I do not believe, however, that natural vascular grafts have been definitely ousted from vascular surgery; one is rather waiting for some new discoveries in angiology which could be applied for the improvement of the results obtained with the natural vascular grafts as could be seen in the revival of some vascular grafts (3). But even then the natural vascular grafts will not be able to compete with alloplastic prostheses in their availability and easy preparation.

The Tissue Bank should always keep preserved tissues of various sizes and shapes ready for use. The best evidence that the morphological adjustment of the graft will shorten the operation and make the surgeon's work easier is offered by the preparation of a preserved foreign graft for osteosynthesis of the spine by the method of Cloward (4). Morphological adjustment is also advantageous for the economic manipulation of this precious material. Economic considerations, however, must not cause the surgeon to have too little material for the intended operation because in this way one of the main

*) Paper read before the International College of Surgeons, Vienna, May, 1964.

advantages of the use of preserved tissues, namely the independence of the donor, would be lost.

Since every tissue and consequently every kind of graft has its specific physiological function which need not necessarily be congruous with the intended application (especially if this application is heterotopical), it is necessary to withdraw a suitable tissue and to adjust and preserve it properly. For example if we want a cartilaginous graft to be the most suitable substrate for bone rebuilding in an osteomyelitic cavity we remove the perichondrium and cut the material into small pieces which can completely fill all sinuses (5). On the contrary, in grafts intended for plastic operations of the nose we carefully remove only the neighbouring tissues trying to preserve the perichondrium as intact as possible, and then we plunge the graft into a warm bath to decrease its elasticity so that it will keep its shape better (6). A good preservation is not easy because in its course we change the natural conditions of the existence of the tissue, which have developed in vivo for a long time and we replace them by new conditions intended to be most suitable for the preservation of a certain state of the tissue in vitro. This task is most difficult during hemibiotic preservation. In preservation we use the effect of cold, drying and, sometimes of chemical changes in the environment. It is obvious that our treatment which affects the delicate and complicated structure of a living substance, is very coarse and, consequently, that the procedures fulfil the demands on the properties of the tissues to a different degree. Thus, if all the known factors in the graft are not taken into account the result of the operation is not attained from all aspects and failure may occur even if the operation was exactly indicated, technically faultlessly performed and if the postoperative course was uneventful. On the other hand, we must not overestimate the value of a good graft either, for it is not a jolly joker capable of solving everything; we must always keep in mind that each graft has its limited indications. Thus, e.g., our fascial grafts have been successfully used in plastic operations of dura mater and, what is especially important, even in cases complicated with liquorrhoea, in various hernioplasties, especially in relapsing conditions in aged patients, and also in an operation for Morgani's diaphragmatic hernia, or as fillings in the operation for retinal detachment, in plastic operation of the ligamentum patellae proprium and in the reconstruction of the tendon of the m. rectus femoris. They failed, however, in tympanoplasties and, as ultimum refugium in a case of gastroschisis with a complete defect of the anterior abdominal wall and congenital peritonitis when nearly all the organs of the abdominal cavity were exposed and the skin of the mother was not available.

A systematic long-term control of the subjects operated on, in which the worker of the Tissue Bank takes also part, is the beginning of the second task — research. The final evaluation of the indispensable laboratory studies and experiments on animals, depends of course, on the clinical results. The experimental results must be evaluated with caution for clinical application, as we have ourselves confirmed [e.g. in freeze-dried calf pituitaries used in a complex treatment of perinatal encephalopathy (8)]. Research is an un-

separable part of any progressive medical study and the development of transplantations is unthinkable without an intensive basic and applied investigation of a new paraclinical line — the preservation of tissues. This is why research is an unseparable part of the activities of the Tissue Bank. Our knowledge of the properties of the grafts, indispensable for successful transplantation, is very incomplete. This condition is no longer tenable, since we must consider the graft to be a pharmacological agent. The fact is that we do not treat the patient with five tablets a day or by an injection but by an exactly known amount of a certain substance administered in five doses a day or in an injection containing a certain amount of the effective drug in water solution. Our present knowledge of grafts shows that, in a majority of cases, we apply a tissue which is morphologically normal, and we also know whether it is sterile or not. Surgeons using freeze-dried bones, may be interested in our communication reporting how the time of rehydration essentially changes the biochemical composition and mechanical properties of these tissues (9). The gaps in our knowledge are also related to certain basic questions, e.g. the indication and contraindication of taking grafts, some changes occurring during treatment and preservation, and the question of expiry which is closely connected with them. In practice, some of these questions are solved on the basis of observations derived from isolated, unclear cases, though some have already been cleared up or disproved.

We shall also have to turn our attention to the effect of therapy, especially long-term therapy, on the properties of tissues. The assumption that the donor should be a young healthy individual who died following an injury, may appear completely misleading. We must realize that only a few individuals die immediately after the injury and, consequently, traumatic shock develops in the others, which deeply influences the metabolism and thus also the composition of the tissue. The more intensive is the metabolism of the tissue, the deeper are these changes. In general it is stated that bradytrophic tissues can be successfully transplanted. Modern physiology shows, however, that even tissues with such poor cellularity as can be found in the cornea and the cartilage have a relatively high metabolism. Even bone cannot be considered as a bradytrophic tissue because 30% of its dry-weight, which consists of organic substances, has a very intensive metabolism. We try to control the shock therapeutically and thus other factors appear on the scene which act in a complex manner in the organism interfering also in relations other than those for which they were used. Thus, in 63% of our donors antibiotics (24% of them being broad-spectrum antibiotics) were administered before death. The administration of antibiotics, chemotherapeutics, hormones and certainly also of other drugs, especially if they are given in large doses and for a long time, will not remain completely without influence on metabolism (10). These changes may be especially manifest in tissues preserved hemibiotically or cultivated in vitro. On the basis of a few observations we have the impression that transfusions, and thus perhaps even serotherapy and immunotherapy, influence the homotransplantation reaction (11). Further effective factors

assert themselves with the gradually worsening harmony of the vital processes passing through to the stage of clinical death, necrobiosis, which finally affects practically any tissue withdrawn after death. This results in the fact that only in a few isolated cases do we obtain — sit venia verbo — a "normal" tissue while in most of the tissues used there is considerable variability in the properties of the individual tissues from different donors.

It may appear that such contemplations are sterile and that the use of fresh tissues would be the best solution. Clinical practice based on a large number of well known reasons and cases shows that, in most cases, this method is not convenient. But if we demand from the transplants more than the basic physical functions — and these demands can be fulfilled by the natural grafts in contrast to alloplastic prostheses — then this criticism is justified. This is established by the fact that the investigation of hemibiotically preserved grafts can be carried out up to the physiological level whereas the investigation of abiotic and anabiotic grafts at the biochemical level at the utmost, and the study of alloplastic prostheses only at physical and chemical levels.

In my opinion, the study of the changes in the antigenicity of the grafts and the related question of the possibility of using heterogenous grafts in clinical practice is the most important task. By the reduction of antigenicity of the grafts I unterstand a qualitative change in the transplantation reaction which may also manifest itself by a reduced number of recipients in whom we shall be able to observe a certain kind of the immunological reaction. Thus, e.g., after the application of heterogenous bone grafts processed according to the Czechoslovak patent No. 92515, which has been elaborated in our laboratory, we have been able to demonstrate the development of humoral antibodies of the ABO system in 64% of recipients, and an increase in gama globulins in 45% of recipients, as compared with the anticipated 100% after the application of fresh unpreserved heterogenous grafts, and 50% of ABO antibodies and 30% increase in gamma-globulins after homogenous keratoplasty, which is associated with the mildest homotransplantation reaction. The application of a graft with a reduced antigenicity is doubtlessly a better procedure than the influencing of the transplantation reaction by treating the recipient, since the present methods designed to inhibit this reaction simultaneously retard the regeneration.

Extremely important for the formulation of the research problems — sometimes partial, sometimes of basic importance — are cases of unexpected clinical success, and especially failure — among them, in the first place, the so-called diseases of the graft. Their exact analysis and recording leads to very useful conclusions which can immediately be used in practice. It is necessary that a worker from the Tissue Bank should take part in the evaluation of these cases also, because in this way the problem can be solved on a broad basis using the experience of other departments and with other tissues. He alone as an expert in transplantation can be familiar with such detailed knowledge. In the first place the worker at the Tissue Bank must express his own

views about the properties of the graft and some relations between the graft and the recipient. Cooperation between the clinical departments and the Tissue Bank must not be limited to the filling-in of forms but it must be a personal and reciprocal cooperation and it must be kept in mind that the availability of preserved tissues and transplantation "fashions" should not lead to its discredit.

The third task of the Tissue Bank is education which, affording ready information on new advances contributes to their rapid application in practice, and in this way to the improvement of clinical results. Successful postgraduate training of this kind secures a good overall standard which depends on the knowledge of the participants and on their experience, and on their interest in this work. Training is directed, in the first place, to Tissue Bank workers of all categories. Whenever possible lectures should be connected with practical training. This is exceedingly important. In addition, the Tissue Bank encourages special training for clinical workers which is carried out in cooperation with orthopedic, plastic surgeons, or ophthalmologists, who are becoming increasingly interested in transplantations in their fields and studying the basic biological problems of transplantation. The training is either separate or it may be included in the programme of Postgraduate Courses.

In the countries with planned health services the Tissue Bank is also charged with the respective organizational tasks.

For this reason the Tissue Bank occupies a key position in the question of tissue transplantation. Its specialization is a condition sine qua non for the preparation of high quality preserved tissues. It is a natural research centre because it is in close contact with all work going on in transplantation. Since transplantation is a physiological, surgical, therapeutical method and in case of the use of foreign tissues also a kind of — sit venia verbo — prevention, it is finding increasing application in all branches of surgery, and consequently the tasks of the Tissue Bank are steadily increasing.

SUMMARY

The tasks of a Tissue Bank for clinical transplantation are as follows:

1. preparation and distribution of preserved tissues, 2. research, 3. education, 4. organization.

In the part dealing with the first task examples of preparation of various preserved tissues, and difficulties met with in some methods of preservation are mentioned. Laboratory work and experiments on animals must go before the clinical research, the long results of which determine the final evaluation. In this part some important unknown factors are pointed out which influence especially the properties of vital grafts. In the author's opinion the most important research task of the Tissue Bank is the study of the ways for reducing the antigenic properties of the grafts. Special education is planned for Tissue Bank workers of all categories and for clinicians of those branches which are concerned with transplantations. The organization tasks depend on the structure of the health services of the country.

RÉSUMÉ

Le rôle de la Banque des Tissus dans la clinique des transplants

R. Klen

Le rôle de la banque des tissus à l'égard des transplants cliniques est suivant: 1. la préparation et conservation des tissus en question, 2. les recherches, 3. l'éducation, 4. l'organisation.

Dans la première partie, touchant la préparation et conservation des tissus, l'auteur donne l'example des préparations variées des tissus divers, et en indique les difficultés. Les recherches cliniques eux-mêmes doivent être précédées par des examens exécutés sur des animaux et par des examents laboratoires. Seuls les résultats permanents des deux catégories déterminent l'évaluation définitive. Dans cette partie, l'auteur décrit des faits jusqu'alors inconnus qui influencent d'une manière spéciale les qualités des transplants vivants à l'avis de l'auteur, les recherches les plus importantes de la Banque des Tissus, c'est sont ceux des moyens diminuants les qualités antigèniques du transplant. L'auteur donne l'exemple du plan d'éducation des travailleurs de tout grade d'une Banque de Tissu ainsi que des cliniciens de toute branche médicale se servant des transplants. Quand'à l'organisation, elle est due et dépend de la structure elle-même des services de santé du pays.

ZUSAMMENFASSUNG

Aufgaben der Gewebebank bei klinischen Transplantationen

R. Klen

Die Gewebebank hat bei klinischen Transplantationen folgende Aufgaben zu erfüllen:

1. Herstellung und Distribution konservierter Gewebe, 2. Forschungsaufgaben, 3. erzieherische Aufgaben, 4. organisatorische Aufgaben.

Inbezug auf die erste Aufgabe bringt die vorliegende Arbeit Beispiele für die Herstellung verschiedener konservierter Gewebe sowie für die Schwierigkeiten bei manchen Konservierungsmethoden. Laboratoriumsuntersuchungen und Tierversuche müssen der klinischen Forschungsarbeit vorangehen, die Spätergebnisse der letzteren sind für die endgültige Bewertung massgebend. In diesem Teil der Arbeit werden manche wichtige, jedoch wenig bekannte Faktoren hervorgehoben, die namentlich die Eigenschaften lebender Transplantate beeinflussen. Nach der Ansicht des Verfassers besteht die wichtigste Forschungsaufgabe der Gewebebank im Studium von Methoden, mittels deren die antigenen Eigenschaften der Transplantate reduziert werden können. Eine spezielle Ausbildung wird für alle Mitarbeiter der Gewebebank aller Kategorien geplant, ebenso für Kliniker derjenigen Fächer, in denen Transplantationen in Frage kommen. Die organisatorischen Aufgaben hängen von der Struktur des staatlichen Gesundheitsdienstes ab.

RESUMEN

Los trabajos del Banco de Tejido en las transplantaciones clínicas

R. Klen

Los trabajos del Banco de Tejido para la transplantación clínica, son los siguientes: 1. preparación y distribución de los tejidos preservados, 2. investigación, 3. educación, 4. organización.

En la parte que trata sobre el primer trabajo, se mencionan los ejemplos de la preparación de varios tejidos preservados y las dificultades reunidas en algunos métodos de preservación. El trabajo de Laboratorio y los experimentos con animales deben preceder a la investigación clínica, los extensos resultados de los cuales, determinan la evaluación final. En esta parte se ponen de manifiesto algunos factores desconocidos de gran importancia, los cuales influyen especialmente sobre las propiedades de los injertos vitales. En opinión del autor, el más importante trabajo de investigación del Banco de Tejido, es el estudio de las formas de reducción de las propiedades antigénicas de los injertos. Una educación especial está planificada para los trabajadores de todas las categorías del Banco de Tejido, y para los clínicos de las ramas, que son concernientes con la transplantación. Los trabajos organizativos dependen de la estructura de los servicios de salud del país.

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HOMOTRANSPLANTATION OF ARTERIES BASED ON TOTAL BLOOD EXCHANGE TO OVERCOME TISSUE INCOMPATIBILITY

(Experimental Study)

M. S. ZNAMENSKY, T. S. ODARYUK

The phenomenon of immunological tolerance, described by Hašek, Medawar and Puza in animals during the embryonic and postnatal period, inevitably raises the question of whether similar phenomena can be produced in adults, since it is evident that clinical application depends on the solution of this question.

Some experiments along these lines have already been carried out, but they did not produce any promising results. Puza and Gombos carried out homotransplantation of skin in dogs using exsanguination-transfusion one month after grafting, but failed in adult animals.

The experiments of Chepov, Puza et al. with homotransplantation of the kidney after two successive total blood exchanges, were also unsuccessful. They carried out a total of four experiments and in all of them the animals died after a short period.

On the other hand, it was known that homotransplants of tendons, cartilage and bone tissue, cornea and blood vessels have taken successfully. It thus becomes evident that the various tissues possess different antigen levels.

In view of these facts we attempted homotransplantation of an arteries, trying to overcome tissue incompatibility by total exchange transfusion.

MATERIAL AND METHOD

Large non-pedigree dogs were used as experimental animals. A total of four series of experiments were carried out. Pairs of dogs of the same sex and — if possible — the same weight were selected as recipient and donor. All operations were carried out under local anaesthesia. The suture between the graft and the vessels was performed by the vessel suturing apparatus in all cases.

Series I: Homotransplantation of the abdominal aorta carried out in 20 dogs. In the donor dog the femoral artery was exposed, heparin injected and after five minutes the animal exsanguined through that artery until the cessation of blood flow (usually 800 to 1500 ml of blood was obtained). Then a section of the abdominal aorta, 6—8 cm. in length, was excised and submerged in a vessel filled with the heparinized blood.

The abdominal aorta of the recipient dog was exposed and a section of 2—5 cm. in length excised between the origins of the renal and the inferior mesenteric arteries. The defect was then bridged by the transplant which was fixed to both stumps end-to-end by vessel suture. The animal was then exsanguinated through the femoral artery and blood replaced by an equal amount of donor blood.

Glozman and Kassatkina, who first described total exchange transfusion, call this method "the 100% exchange" (relatively speaking, since some of the original blood remains in the recipient organism). In the postoperative period two dogs died from haemorrhage through the vessel suture due to a technical error during operation. One dog died from peritonitis which developed due to necrosis of the intestine resulting from the ligature of the inferior mesenteric artery carried out by mistake. The remaining dogs remained well and good pulse developed in their femoral arteries. They were kept under observation for 804 days. Autopsy demonstrated well patent transplants.

Series II: Homotransplantation of abdominal aorta combined with a 200% exchange transfusion. The donor blood and aortal transplant were obtained by the above described method. In the recipient dog a section of aorta was replaced by the homotransplant. The dog was then bled from the femoral artery in an amount corresponding to that of the blood obtained from the donor dog, but this amount was first replaced by saline. This was followed by the intravenous infusion of the donor blood while the animal continued to be bled from the femoral artery. This procedure has been tentatively called 200% blood exchange. A total of 19 dogs was used in this experiment. They were kept under observation for 787 days. In two cases haemorrhage occurred at the site of the anastomosis. One dog died from overdosage of heparin.

Series III: Homotransplantation of abdominal aorta combined with two successive 200% blood exchanges at an interval of five days. A total of 10 dogs was used in this experiment. In order to save the donor for the second exchange transfusion, the blood taken from it was replaced by the blood of the recipient.

Controls: In ten dogs homotransplantation of the abdominal aorta was carried out without exchange transfusion: in two of these, thrombosis of the aorta developed and in one the vessel suture ruptured. In one dog a severe scar stricture of the aorta developed after 154 days. The remaining six dogs were kept under observation for 280 days.

Series IV: Homotransplantation of the common carotid artery after double exchange transfusion between the donor and recipient dogs. In the first

exchange transfusion the femoral artery of one dog was connected up with the femoral vein of the other and vice versa and the pooled circulation continued for 1 to 2 hours. Five days later an analogous procedure was carried out from the carotid artery into the jugular vein of the two dogs. Then a section of the carotid artery from the other side was excised and cross-transplanted. One dog of this series died. Out of the 19 experimental animals 12 developed a fully patent transplant, in seven thrombosis occurred.

In the control group, eight out of ten dogs developed thrombosis and in two the patency of the transplant was preserved.

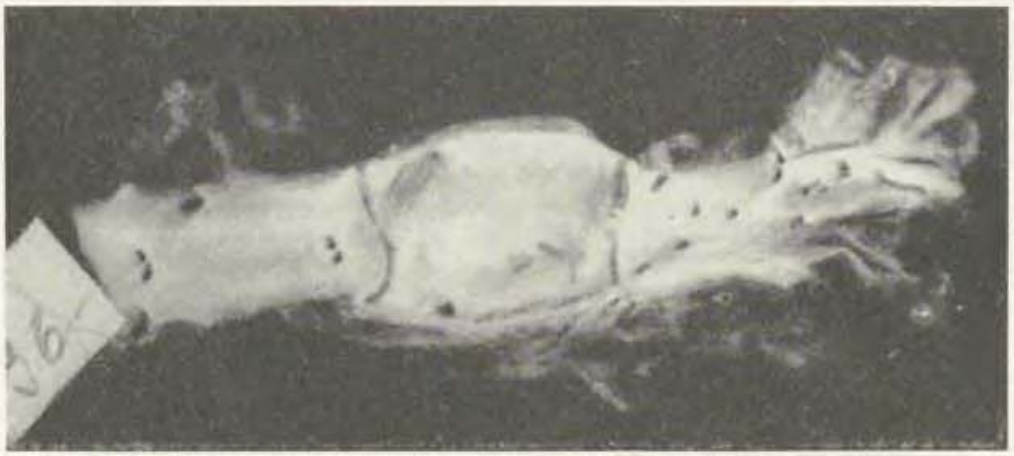

Fig. 1. Homotransplant of abdominal aorta 804 days after implantation and 100% exchange transfusion.

DISCUSSION

Following up the fate of the homotransplant, certain features were observed indicating a slowly developing process of transformation in the homotransplant. Histological changes were seen at two sites of the specimen, i.e. at the proximal vessel suture and in the middle section of the transplant.*)

The stump of the recipient's vessel, taking part in the anastomosis, did not suffer any gross changes; in the early stages changes consisted only in some oedema and slight loosening. The cuff of the vessel end turned up over the plug of the vessel suturing device, showed an absence of cells and structure. However, elastic fibres were preserved in it up to the time of observation.

Between the arterial wall and the transplant, in a small groove at the site of the flexion, a mural wedge-shaped thrombus developed which smoothed out the uneveness of the vessel wall. The fibrin clot finally became organized, with subsequent scar formation. Endothelial cells from host artery spread over its surface and that of the transplant. Much later fine elastic fibres appeared in the wedge-shaped scar. Later still even smooth muscle cells could be detected (Fig. 2).

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*) All specimens were examined by Prof. B. F. Malyshev, for which we wish to express our sincere thanks.

Fig. 2. Wedge-shaped scar between transplant and aorta of host 804 days after operation. Scar contains dense network of elastic fibres. Holes left after removal of clamps. Magnified 80×, stained with resorcin-fuchsin.

Fig. 3. Homotransplant of abdominal aorta 268 days after implantation. Dense elastic network in intima and media. Magnified 330×, stained with resorcin-fuchsin.

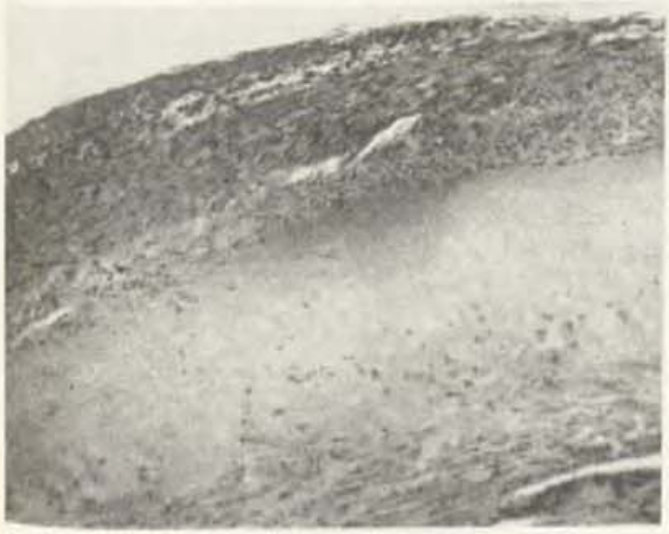

Fig. 4. Control: Homotransplant of abdominal aorta 154 days after operation. Intimal thickening due to fibroblast infiltration. Media without structure. Angiogram: marked stricture of lumen of transplant.

2. In the middle section of the transplant, necrosis and desquamation of endothelial cells of the intima were observed on the 9th to 13th day. Afterwards the endothelial cover was renewed by the endothelium growing over the intima of the graft from the lining of the host's artery. The fibre structure was preserved as proved by staining elastic tissue. The elastic fibres stained well even after 804 days. Already at an early stage the fibre network was infiltrated by fibroblasts of the host which led to thickening of the intima. Starting on the sixth to eighth day newly developed capillaries and nerve fibres invaded the intima from the surrounding tissue (Fig. 3).

The media preserved its muscular structure for a long time, but the thickness of the muscle layer gradually diminished, mainly at the expense of the peripheral part. Later still almost the entire muscle layer was replaced by fibroblasts, although occasional muscle cells were preserved up to the end of observation (804th day). In some specimens new smooth muscle cells were found in the deep layers of the intima. The elastic matrix of the media was preserved and could be demonstrated at all stages of observation. In the later stages, a well distinguished elastic membrane was detected in some specimens.

The adventitia of the transplant mainly underwent fibrous transformation At an early stage newly developed blood capillaries and nerve fibres, penetrating from the surrounding tissue, made their appearance and invaded the media and intima.

At the latest period of observation elastic fibres were detected in the adventitia.

In the controls the picture was dominated by the destruction of elastic fibres in addition to cell elements. All tissues were finally replaced by fibrous tissue (Fig. 4).

An analysis of histological findings shows that all cells of the transplant without exception underwent necrosis; the endothelium of the intima at an early stage, the muscle cells much later. The endothelium regenerated at an early stage from the artery stump of the host. Muscle cells were replaced by fibroblasts. After a 200% blood exchange the muscle cells were more stable and could be seen even after 787 days (the latest time of observation). Two successive 200% blood exchanges did not improve the results.

An analogous fate of the homotransplant was also observed by other authors as, for example Abdyldayev, after implantation of a skin homotransplant under the skin of the rabbit ear, Artomonova after homotransplantation of the tendon graft in the repair of a tendo Achillis defect, Krupko and Tkachenko, and Kovalenko and Zvonkov after repair of bone defects with stored bone homotransplants.

Ratner carried out arterioplasty using stored homotransplants. On histological examination he found necrosis of the intima endothelium (which he ascribed to the surgical trauma) and mural thrombi at different places already in the first few days after operation.

In her experimental study dealing with the same problem, Barsova maintains that the endothelial cells already undergo necrosis on freeze preser-

vation. Subsequently fibroblasts, growing from the aorta intima, form the cover of the inner surface of the transplant (our findings show that these fibroblasts are actually endothelial cells). The newly formed intima thickens through infiltration of cells with elongated nuclei.

Lyophilized transplants are prone to the destruction of fibrous structures in the wall with the subsequent development of aneurysms and even rupture (Petrovsky).

All these examples demonstrate that the cell elements undergo necrosis while the fibrous structures are preserved. The subsequent invasion by cells from the local tissue forms a "hybrid tissue" of a special type.

All the above authors see no basic difference in the fate of a homo- or autotransplant. Repair of an arterial defect with a homotransplant of a vein, for example, also resulted in necrosis of the endothelium by the eighth to ninth day as shown by the experiments of Eshimbetov (carried out at our laboratories). The endothelium then regenerates, but the intima thickens due to fibroblast infiltration. Another important feature found by this author was the appearance of a massive elastic membrane in the surrounding tissues.

The facts given above show that homotransplants suffer the same fate as autotransplants, i.e. they undergo t r a n s f o r m a t i o n and a s s i m i l a t i o n at the new site. But this can take place only if tissue incompatibility is lowered. In this respect we consider the method of total exchange transfusion, which permits transplantation of living arterial tissue without preliminary preparation, to have a better perspective than the use of lyophilized grafts which are actually but dead organs and serve only as a scaffold for the ingrowing tissues of the host.

The contrasting fate of transplants in the control groups, i.e. tendency to thrombosis, necrosis of cells, the important elements of the vascular wall, the destruction of the elastic tissue and the substitution by fibrous tissue with subsequent shrinkage, confirms the value of our method of overcoming tissue incompatibility.

CONCLUSION

1. An arterial homotransplant implanted into an arterial defect of the recipient integrates with the stump end of the artery by a mural scar and thus serves as a channel for the uninterrupted blood flow.

2. This is possible under conditions where tissue incompatibility has been overcome by the total exchange of blood of the recipient for the blood of the donor. A 200% exchange transfusion is still more effective. Two subsequent 200% blood exchanges do not improve the results.

3. Cross homotransplantation after repeated exchange transfusion gave satisfactory results in carotid artery plasty.

4. Control experiments, carried out without exchange transfusion, were accompanied by severe destructive changes in the tissues of the transplant making it unsuitable for function.

5. A homotransplant implanted into an arterial defect undergoes a long and complicated process of transformation consisting in necrosis and regeneration of endothelium of the intima, necrobiosis of muscle cells and preservation of fibrous structures which are subsequently infiltrated by fibroblasts of the host.

RÉSUMÉ

Homoeoplastie artérielle à l'aide du remplacement total du sang servi de méthode d'omission d'incompatibilité des tissus

M. S. Znamensky, T. S. Odaryuk

1. Homoeogreffe artérielle, placée dans le défect de l'artérie du récepteur, fait l'union avec les deux bouts de cette artérie et sert bien à faire transporter le sang.

2. Ce n'est possible qu'à l'aide d'omission de l'incompatibilité des tissus par la méthode de remplacement du sang du récepteur par celui du donneur. Encore plus d'effect donne le remplacement total à 200%. Pourtant celui-ci répété ne montre aucune augmentation du succès.

3. Homoeogreffe croisée de l'artère carotide pratiquée à l'aide du remplacement croisé répété a été très effective.

4. Les contrôles faites sans le remplacement du sang ont été suivies par des changements déstructifs qui causèrent son fonctionnement difficile et presque impossible.

5. Homoeogreffe placée dans le défect artériel est soumise au proces très compliqué et difficil de transformation: la déstruction et réconstruction de l'épithélium de la partie intime, la déstruction des différentes nobles structures des tissus et leur remplacement par les fibroblastes du récepteur.

ZUSAMMENFASSUNG

Homotransplantation einer Arterie bei totalem Blutaustausch, um Gewebeinkompatibilität zu vermeiden (Experimentelle Studie)

M. S. Snamenskij, T. S. Odaryuk

1. Ein arterielles Homotransplantat, in einen Arteriendefekt des Empfangers eingesetzt, verwächst mit dem Arterienstumpf unter Bildung einer Wandnarbe und dient so als Kanal für den ununterbrochenen Blutstrom.

2. Dies wird dadurch ermöglicht, dass die Gewebeinkompatibilität durch totalen Blutaustausch vermieden wird, wobei das Blut des Empfängers durch das des Spenders ersetzt wird. Eine 200%-Austauschtransfusionen ist noch wirksamer. Zwei aufeinander folgende 200%-Austauschtransfusionen verbessern die Ergebnisse nicht mehr.

3. Kreuzweise Homotransplantation nach wiederholten Austauschtransfusionen ergab bei Plastik der Arteria carotis zufriedenstellende Ergebnisse.

4. Kontrollversuche, die ohne Austauschtransfusion verliefen, ergaben schwere destruktive Veränderungen im Gewebe des Transplantats, wodurch es seine Funktion nicht mehr erfüllen konnte.

5. Das in einen Arteriendefekt transplantierte Homotransplantat unterliegt einem lange dauernden und komplizierten Umwandlungsprozess, der in Nekrose und Regeneration des Intima-Endothels, Nekrobiose der Muskelzellen unter Erhaltenbleiben der bindegewebigen Strukturen besteht; die letzteren werden sodann von Fibroblasten des Empfangers infiltriert.

RESUMEN

La homotransplantación de la arteria, basada en el cambio total de la sangre, para superar la incompatibilidad tisular (Estudio experimental)

M. S. Znamenskij, T. S. Odaryuk

1. Un homotransplante arterial implantado dentro de un defecto arterial del recipiente, se integra a la terminación de muñón de la arteria por una cicatriz mural y de este modo sirve como un canal para el flujo ininterrumpido de la sangre.

2. Esto es posible bajo condiciones donde la incompatibilidad tisular ha sido superada por el cambio total de la sangre del recipiente para la sangre del donador. Un 200 % de cambio en la transfusión, es todavía más efectivo. Dos subsiguientes 200 % de cambios transfusivos, no mejora los resultados.

3. Por el contrario, la homotransplantación después de repetidos cambios de sangre, dió resultados satisfactorios en la plástica de la arteria cenótida.

4. Los experimentos de control, llevados a cabo sin cambio transfusivo, estuvieron acompañados de severos cambios destructivos en los tejidos del transplante, haciéndolos inadecuados para su función.

5. Un homotransplante implantado en un defecto arterial, sufre un largo y complicado proceso de transformación, consistente en necrosis y regeneración del endotelio de la íntima, necrobiosis de las células musculares y la preservación de las estructuras fibrosas, las cuales son subsiguientemente infiltradas por los fibroblastos del huésped.

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DETERMINATION OF THE CONDITION OF VASCULARIZATION OF PEDICLE SKIN GRAFTS (GILLIES-FILATOV) BY THE THERMOELEMENT

W. TAUBENFLIEGEL, Z. WAJDA, A. LEWIŃSKI

The most important problem in pedicle skin grafts is the development of the arterial and venous circulation which determines the viability. The development of the blood supply in a pedicle flap is based on the normal time of healing which clinical experience shows to vary from 20—35 days. Uncertainly as to the viability of a flap results in prolonging hospitalisation and the delay of further operations. Several tests have been introduced to determine the exact state of vascularization of pedicle skin grafts, e.g. radioactive sodium

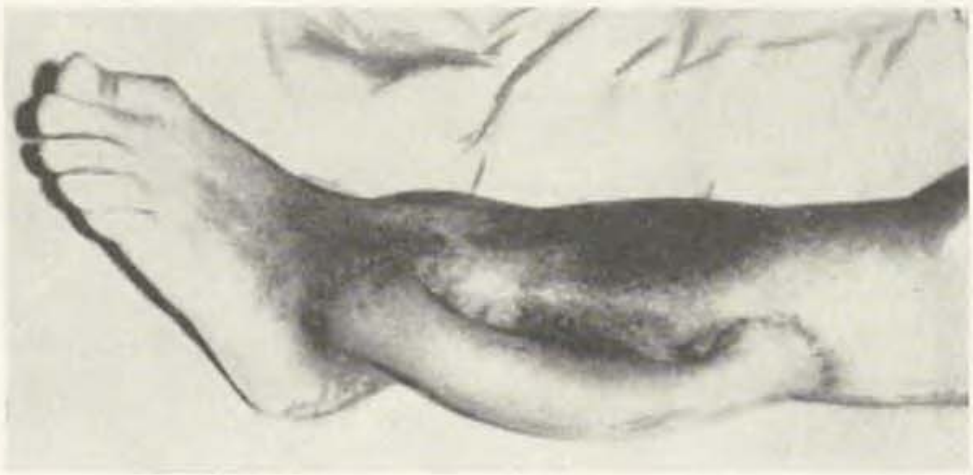

Fig. 1. Photograph of a prepared flap.

chloride and radioactive sodium iodide, the fluoresceine test, X-ray, atropine test, observations with a photoelectric cell for determination of the amount of oxygen in arterial blood and a thermoelectric method introduced by Wust & Bush in 1950.

Assuming that the local temperature in the skin of a flap depends on the blood flow, we made serial tests of the superficial temperature in pedicle flaps at different stages after they had been formed. The thermoelement —

Electric Universal Thermometer type TE 3 was used. Accuracy to 0.1 centigrade.

We investigated two pedicle flaps at different stages after formation (Fig. 1), one being transferred three times from abdomen to thigh, thigh to leg and then to cover the defect. The second flap was transferred twice.

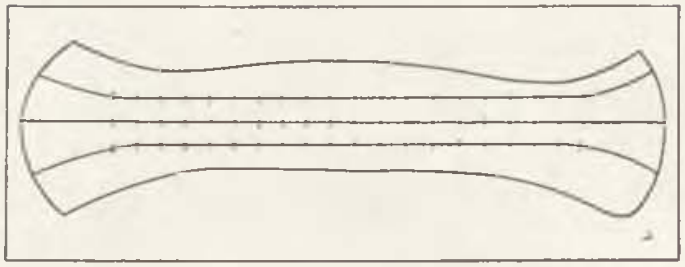

Fig. 2. The measurements along three parallel lines in the long axis of the pedicle at points 1 cm. apart.

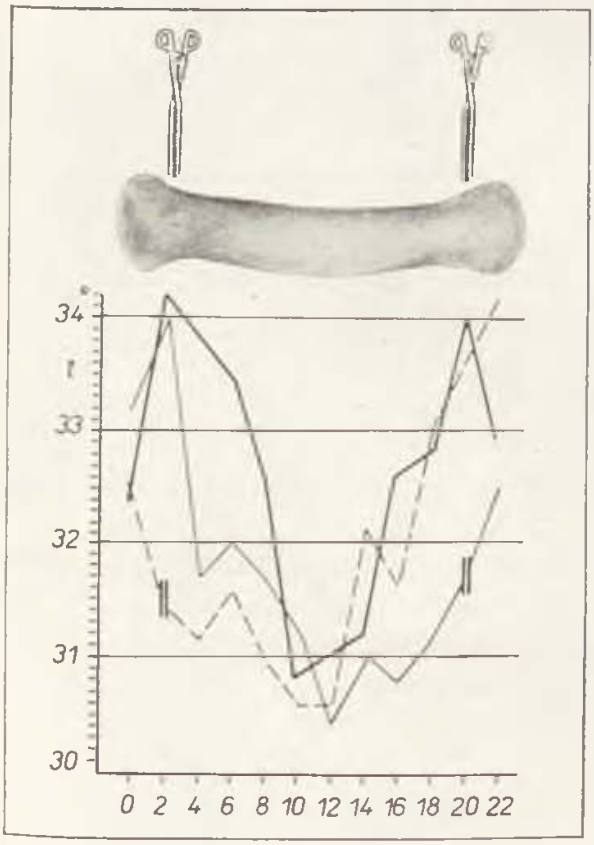

Fig. 3.

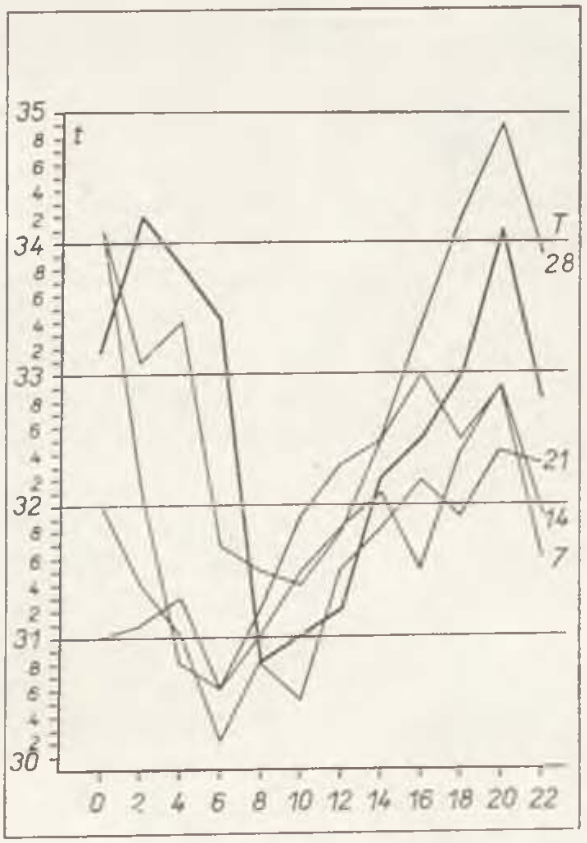

Fig. 4.

Fig. 3. Temperature curves of the pedicle flap 4 weeks after formation (the base-line curve — continuous thick line). The interrupted and thin lines correspond to temperatures after tightening one of the bases of the pedicle. Short vertical lines correspond the sites of compression with a clamp (case 1). *y* — temperature, *x* — centimetre. — Fig. 4. Temperature curves 7, 14, 21 and 28 days after transplantation of one of the bases of the pedicle (The thick line corresponds to the basic temperature curve). y — temperature, x — centimetre, tines — days.

The temperature was measured in both pedicles at definite, constant points, at the time of their formation and at different stages four weeks after formation. The points were 2 cm. from the base of the pedicle and 1 cm apart, along three parallel lines in the long axis of the pedicle (Fig. 2). The average of three values was taken and is given in the figures. The pedicle grafts were

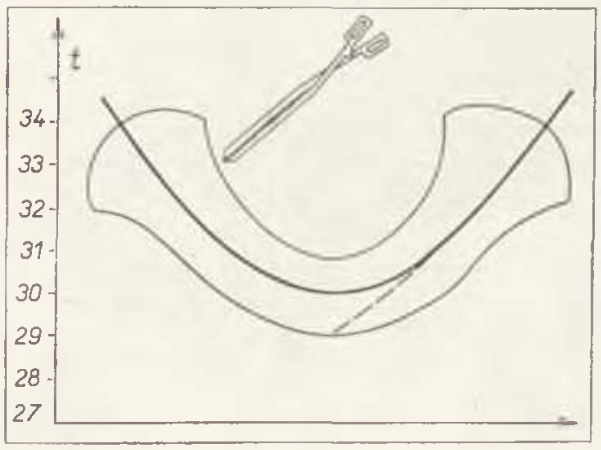

Fig. 5.

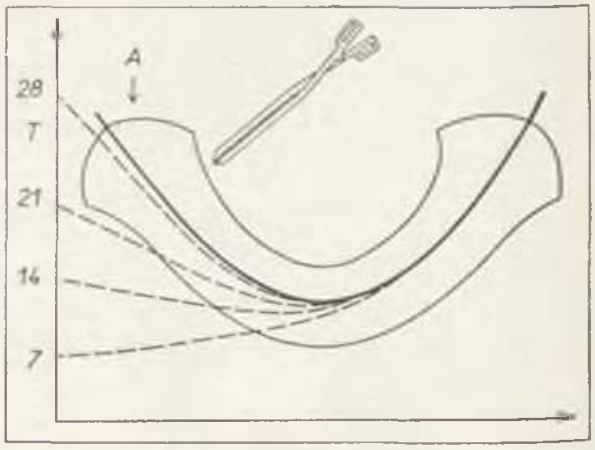

Fig. 6.

Fig. 5. The fall of temperature in the pedicle flap after tightening of one of the bases (interrupted line), 4 weeks after its formation. y — temperature. — Fig. 6. Development of circulation to the transplanted base. Determination of the temperaure curve 7, 14, 21 and 28 days after transplantation of one of the bases of pedicle (case 1). *y* — days, *x* — the base of the pedicle transplanted.

left uncovered at room temperature, all the time, and during the investigations the body temperature of the patients was normal.

The first determination of temperature was usually made 4 weeks after the formation of the pedicle — stage I, and this curve was considered to be the base-line (Fig. 3). This curve had the following characteristics: the highest temperature (on average 34 °C) was at the base of the pedicle. Away from the base the temperature decreased sharply and in the central part of pedicle the temperature was lowest. The fluctuation of the temperature between the central part of the pedicle and the base was 2—3 °C, the lowest point being in the middle of the pedicle.

Investigations made the first day after the transplantation of the flap (this means 4 weeks after its formation) showed that the curve has basically the same characteristics as the curve we got immediately before the transplantation. Further investigations made 7, 14, 21 and 28 days after showed that the curves we got were parallel, but a little lower than the basic one (Fig. 4).

Four weeks after the curve was approximately the same as the one immediately before the transplantation (basic). We considered this period to be the best for further operations. The clinical results confirm the development of an adequate circulation.

We also used the clinical method for determination of the blood flow by gradually compressing the base of the pedicle with clamps (Fig. 5).

This curve has the following characteristics: on compressing one base of the tubed pedicle (i.e. cuting the blood supply on one side) the curve showed distinct decrease of the temperature near the point of compression. The lowest point of this curve was one degree lower than the lowest point in our basic curve. We considered this to be the adequate time for further operations, as confirmed by good clinical results (Fig. 6).

Our next investigations will be concerned with determining whether it is possible to cut the circulation in the pedicle still earlier i.e. when the temperature is lower than 1 °C below that in the basic curve. We are also continuing examination of the vascularity of pedicles immediately after their formation to determine the exact time of the first operation.

CONCLUSIONS

1. The characteristics of the basic curve 4 weeks after the formation of a pedicle skin graft: the curve is similar to a reverse parabola in which the lowest point corresponds to the centre of the pedicle and the highest is at the base.

2. Tightening one of the bases of the pedicle causes a fall of the temperature curve and the lowest point is 1 °C lower than the lowest point in our basic curve.

3. After operating on the pedicle the daily measurements of the temperature gives a curve identical to the one we got after tightening the base. In the course of 4 weeks we could observe that this curve gradually rose and after 4 weeks it is identical with our basic curve. This is the best moment to continue further operations as the vascular network is well developed.

4. By tightening the one intact pedicle that is the source of the whole blood supply for the flap we got a curve similar to the one described under point 2. This confirmed that a good blood supply developed from the new source.

SUMMARY

The authors describe the development and the state of vascularization of the pedicle skin grafts by using the electric thermometer type TE 3. The results were determined on the basis of the superficial temperature of the flap.

RÉSUMÉ

L'évaluation de l'état circulatoire des lambeaux à pédicule (Gillies-Filatov) à l'aide de thermomètre

W. Taubenfliegel, Z. Wajda, A. Lewiński

Les auteurs décrivent l'évolution et l'état circulatoire dans un lambeau à pédicule. Ils ont employé le thermomètre éléctrique TE 3. Les résultats ont été obtenus à la base des données de la température de la surface du lambeau.

ZUSAMMENFASSUNG

Untersuchung der Kreislaufverhältnisse in gestielten Lappen (Gillies-Filatow) mit Hilfe eines Thermoelements

W. Taubenfliegel, Z. Wajda, A. Lewiński

Die Verfasser beschreiben in der vorliegenden Arbeit Entwicklung und Zustand der Gefässe im gestielten Hautlappen. Es wurde das elektrische Thermometer TE 3 verwendet. Die Untersuchungsergebnisse wurden auf Grund der Temperaturmessung an der Lappenoberfläche erzielt.

RESUMEN

La averiguación del estado circulatorio en los lóbulos peciolados (Gillies-Filatov), con la ayuda de una pila calorífica

W. Taubenfliegel, Z. Wajda, A. Lewiński

Los autores describen el desarrollo y las condiciones de los vasos sanguíneos en un lóbulo peciolado de la piel. Utilizaron un termómetro eléctrico TE 3. Los resultados de la investigación, fueron determinados tomando como base la medida de las temperaturas en la superficie del lóbulo.

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(W. Taubenfliegel, M. D.): 3rd Surgical Clinic, Medical Academy, Gdańsk, Poland

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ANNOUNCEMENTS

"The Brazilian Society of Plastic Surgery announces its IVth National Congress, in Pôrto Alegre, Rio Grande do Sul, from October **5 to 6, 1965.**. The scientific program comprises lectures, films and 3 symposia on "Hand Surgery", "Age and Planning of Congenital Deformities' Surgery" and "Plastic Surgery in Dermatology".

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LE DIAGNOSTIC PRÉCOCE DES CANCERS DE LA BOUCHE

V. LENART, I. F. LENART

Aujourd'hui nous nous demandons si nos connaissances sur le problème compliqué et obscure du cancer buccal ont fait pendant les dernières années quelque progrès?

Dans l'intention de se raprocher de ce problème compliqué, nous avons contrôlé systématiquement 100 malades avec des lésions suspectes de muqueuses malpighiennes dans la région maxillofaciale et oropharyngienne pendant plusieurs années, en revoyant périodiquement des frottis et la biopsie (type d'épithélium, la quantité du glycogène dans l'épithélium, la quantité des muccopolysaccharides dans la substance fondamentale de l'épithélium, le comportement des acides désoxyribonucléiques dans les noyaux des cellules épithéliales, la forme et le nombre des nucléoles, l'acide ribonucléique cytoplasmatique, la membrane basale, des mitoses, la quantité des muccopolysaccharides du tissu fibreux, les infiltrats cellulaires, la qualité du tissu de la sous-muqueuse).

Nous avons employé les méthodes suivantes: hématoxyline-éosine, Van Gieson pour le tissu fibreux, P. A. S. réaction pour le glycogène et pour les muccopolysaccharides, la réaction d'après Feulgen pour les acides désoxyribonucléiques, la réaction selon Brachet pour les acides nucléiques et la méthode d'après Gömöri pour le tissu réticulé.

En même temps nous avons observé aussi le protéinogramme avec la chromatographie ordinaire et avec l'immunoelectrophorèse.

Les résultats sont intéressants.

Dans la plupart des cas chez lesquels a pris naissance la cancérisation, nous notions déjà un certain temps avant la cancérisation clinique manifeste quelques échanges histochimiques et biochimiques.

Ce sont:

Accumulation d'acides désoxyribonucléiques dans les noyaux épithéliaux.

Au moins une mitose atypique dans un champ optique chez un agrandissement 800×.

Dans l'évolution de la cancérisation, nous pouvons observer deux changements des nucléoles. Dans la première phase de la cancérisation les nucléoles

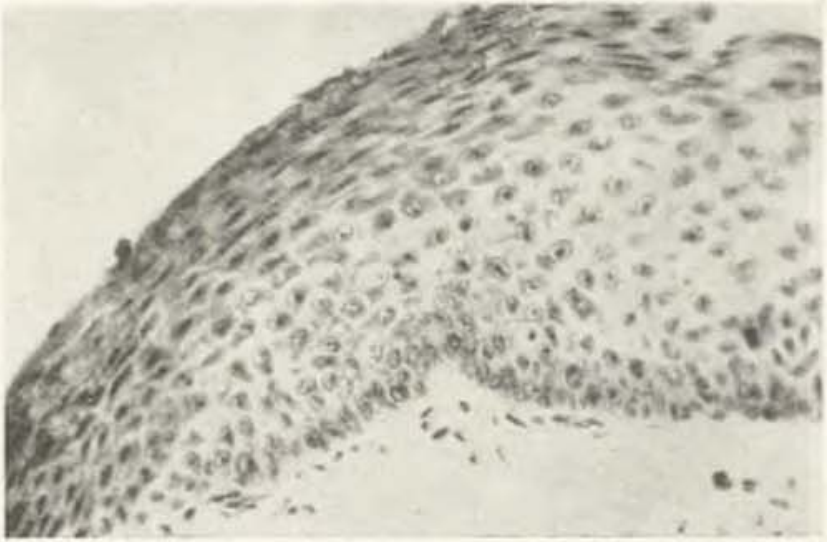

Fig. 1.

Fig. 2.

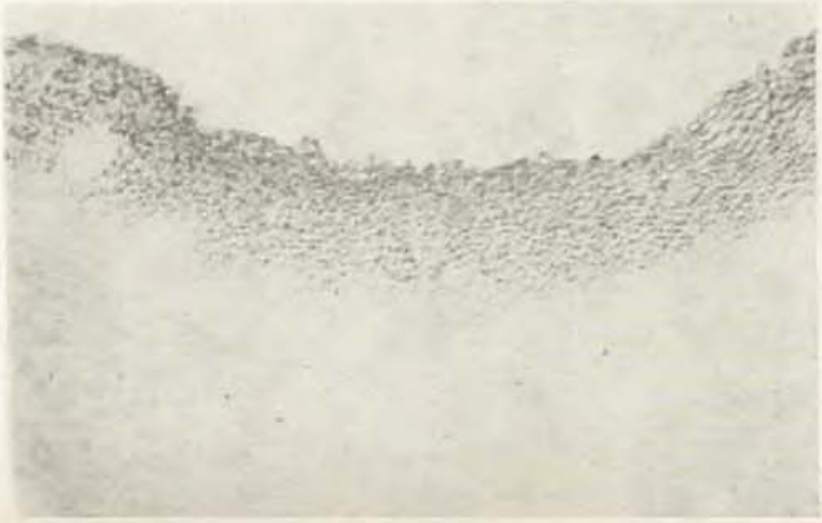

Fig. 1. M. Z. I. Agé 45 ans. Brachet. «Leucoplasie» de la muqueuse malpighienne. — Fig. 2. Même coupe que Fig. 1. Feulgen. La quantité des acides désoxyribonucléiques dans les cellules épithéliales normale. — Fig. 3. Même coupe que Fig. 1. P.A.S. réaction. La distribution de glycogène dans l'épithélium normale. La quantité de la substance fondamentale de l'épithélium et des muccopolysaccharides du tissu fibreux normale. La membrane basale distincte.

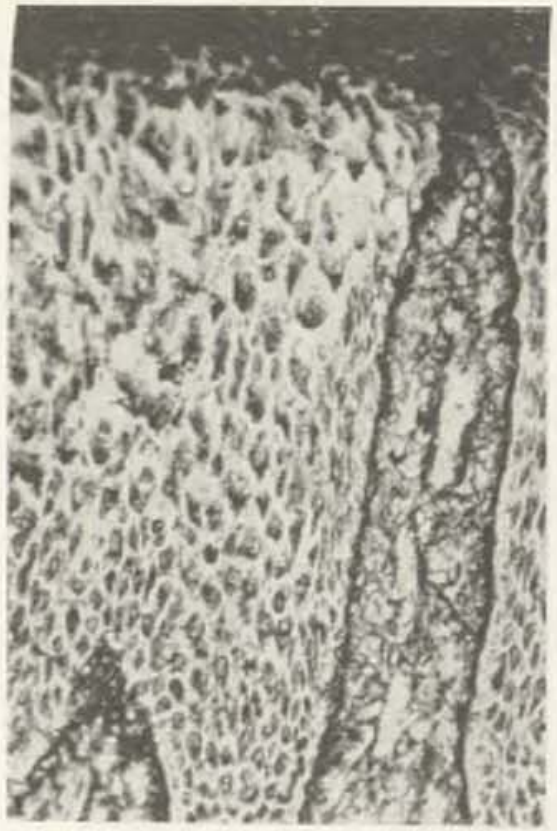

Fig. 4.

Fig. 4. Même coupe que Fig. 1. Gomöri. Il n'y a pas de fibres réticulées parmi les cellules épithéliales. Chez ce cas protéinogramm était normal. Notre premier cas (Fig. 1, 2, 3, 4) a montré le «test de la cancerisation» negatif. — Fig. 5. Mme M.A. Agée 42 années. Brachet. «Leucoplasie» de la muqueuse malpighienne. — Fig. 6. Même coupe que Fig. 5. Une quantité des acides désoxyribonucléiques augmentée. Mitoses pathologiques. Feulgen.

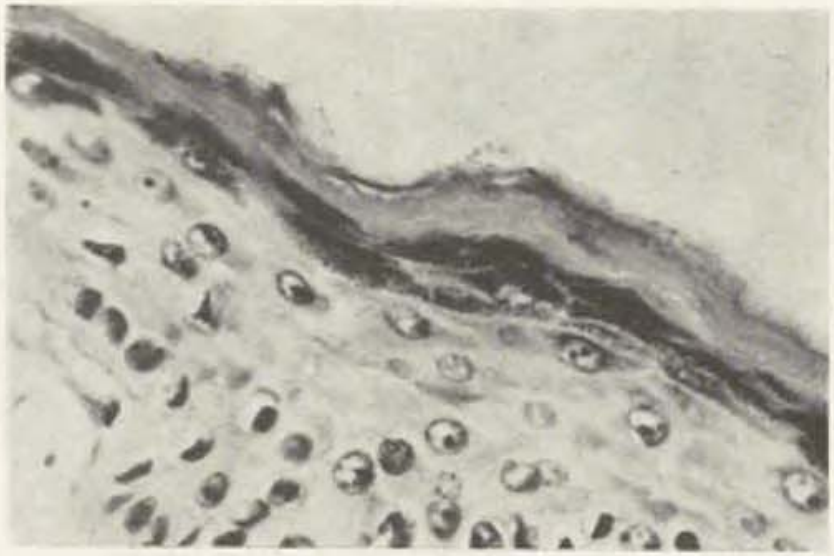

Fig. 5.

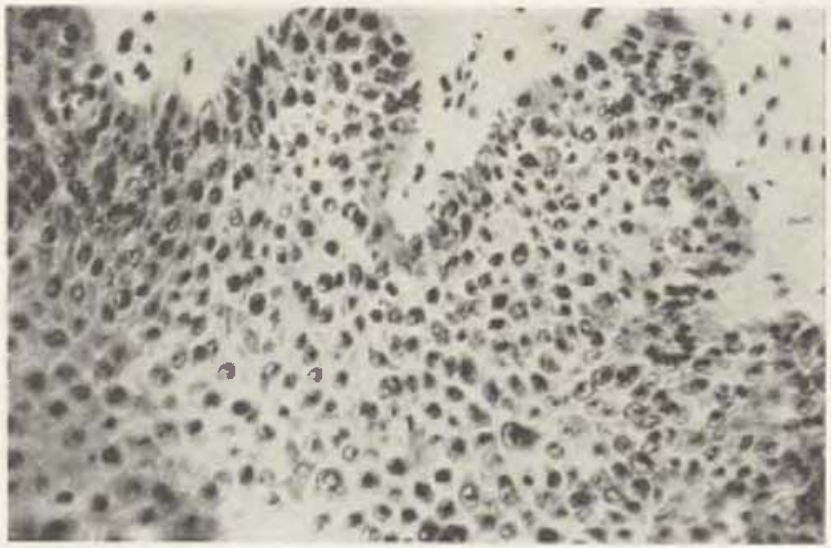

Fig. 6.

Fig. 7.

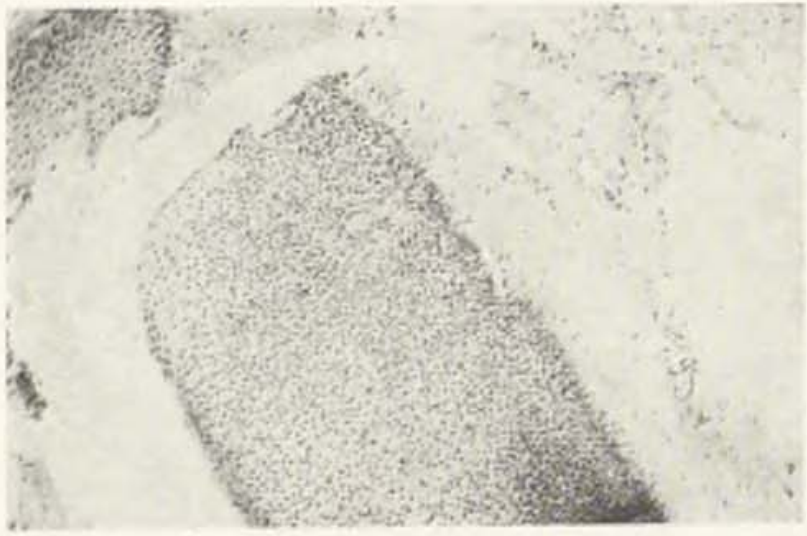

Fig. 8.

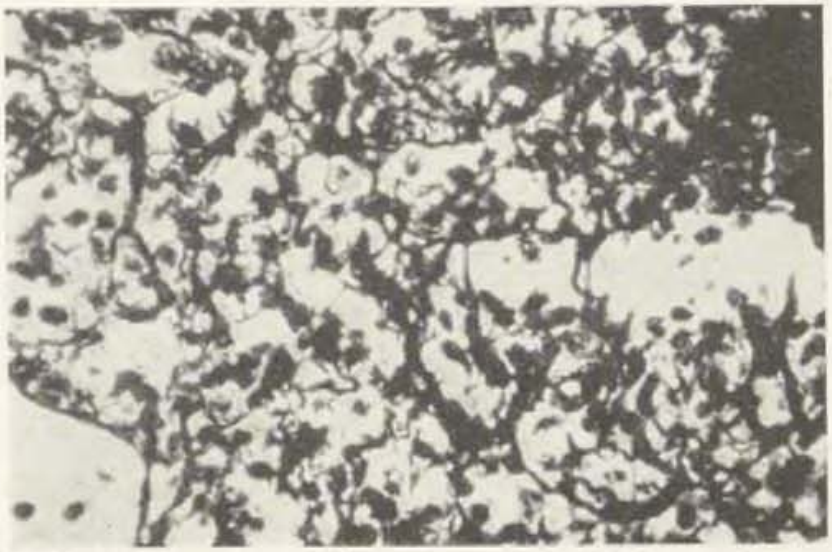

Fig. 9.

Fig. 7. Même coupe que Fig. 5. P.A.S. réaction. Aglycogenie de l'épithélium. La quantité des muccopolysaccharides dans le tissu fibreux augmentée. Une augmentation de la substance fondamentale de l'épithélium. La membrane basale partialément indistincte. — Fig. 8. Même coupe que Fig. 5. Gömöri. L'apparition des fibres réticulées parmi les cellules épithéliales. Chez le deuxième cas le «test de la cancérisation» était positif. — Fig. 9. Même cas que Fig. 5. Après cinq mois une transformation en cancer «In situ». Chez le même cas nous avons aperçu avec immunoelectrophorèse dans le protéinogramme l'apparition des alfa 2, beta 1 et beta 2 globulines.

de l'épithélium sont nombreuses et polymorphes, dans la seconde phase les nucléoles sont minces et moins nombreuses.

Accumulation d'acide ribonucléique cytoplasmatique des cellules épithéliales.

La disparition du glycogène du cytoplasme épithélial.

Accumulation de la substance fondamentale épithéliale.

Apparition de fibres réticulées parmi les cellules épithéliales.

Accumulation des muccopolysaccharides dans le derme muqueux.

Sclerose ou infiltration du derme.

Quant'à la réaction de l'organisme, elle se caractérise par «une crise protéinique» qui peut annoncer la cancérisation définitive quelque temps avant. La crise est caractérisée par une chute des albumines; avec l'immunoelectrophorèse on voit une augmentation des alfa 2, beta 1 et beta 2 globulines. Il est surprenant qu'au moment de la cancérisation manifeste clinique le protéinogramme se normalise quelque fois.

CONCLUSION

Sur la base de nos observations des états précancéreux nous pouvons appréciér la possibilité de malignisation définitive d'une lésion suspecte de la muqueuse malpighienne avec cette fonction mathématique :

$$P = \frac{\text{ADN, ARN, AMPLS, pMI, RSTR, RET}}{\text{GLY}} + \text{RO} + \text{X}.$$

P = Possibilité de la malignisation. — ADN = Accumulation d'acides désoxyribonucléiques. — ARN = Accumulation d'acides ribonucléiques cytoplasmatiques et les atypies des nucléoles. — AMPLS = Accumulation de la substance fondamentale de l'épithélium et des muccopolysaccharides du tissu fibreux. — pMI = Les mitoses pathologiques. — RSTR = Réaction du derme muqueux et de la sousmuqueuse. — RET = Apparition de fibres réticulées parmi les cellules épithéliales. — RO = Réaction de l'organisme. — X = Les autres facteurs (déjà connus et encore inconnus).

RÉSUMÉ

Avec notre conception nous pouvons quelque fois signaler une possibilité d'une malignisation d'un processus suspect. Nous pouvons de même exclure une cancérisation. Ce qui de toute manière présente un grand interêt pour le malade et pour le traitement à apliquer.

Nous savons que nos recherches ne sont qu'un humble pas dans l'obscurité de la question du cancer, mais le pas est fait avec l'espoir de servir.

SUMMARY

Early Diagnosis of Cancer in Oral Cavity

V. Lenart, I. F. Lenart

According to our conception we are, sometimes, able to signalize malignant degeneration of a suspicious process; we can also exclude cancer. This is of great importance to the patient and the plan of treatment.

We are aware of our observations being but a modest contribution to the solution of the problem of cancer. However, the first step has been taken in the hope that it might be helpful.

ZUSAMMENFASSUNG

Frühzeitige Diagnose des Krebses der Mundhöhle

V. Lenart, I. F. Lenart

Auf Grund des beschriebenen Verfahrens sind wir manchmal imstande, maligne Degeneration eines verdächtigen Prozesses zu erkennen oder bösartiges Wachstum auszuschliessen. Dies ist für den Patienten sowie für den Behandlungsplan begreiflicherweise von grosser Wichtigkeit.

Wir sind uns dessen bewusst, dass unsere Beobachtungen nur einen bescheidenen Beitrag zur Lösung des Krebsproblems darstellen. Der erste Schritt ist jedoch getan und es ist zu hoffen, dass er bei der Lösung dieses Problems behilflich sein wird.

RESUMEN

El diagnóstico precoz del cáncer de la boca

V. Lenart, I. F. Lenart

Los autores dan a conocer, que con su concepción, ellos pueden, algunas veces, senalar la posibilidad de una malignización de un proceso sospechoso. Ellos pueden, de la misma manera, excluir una cancerización. Lo que de todas formas presenta un gran interés para la enfermedad y para el tratamiento a aplicar.

Según los autores, ellos saben que los hallazgos no son un simple paso dado en la oscuridad de la cuestión del cáncer, ya que el paso está hecho con el propósito ó con la esperanza de servir.

(Dr. V. Lenart): Česnikova 6, Ljubljana, Yougoslavia

BOOKS

H. J. Denecke, R. Meyer: **Plastische Operationen an Kopf und Hals (in zwei Bänden)**. Erster Band. Korrigierende und rekonstruktive Nasenplastik. — Springer Verlag, Berlin-Göttingen-Heidelberg. — 515 plates (some in colour), XII, and 538 pages (octavo), 1964. Cloth binding, price 328 German marks.

One of the authors, H. J. Denecke, is professor of otorhinolaryngology at Heidelberg, and the other, R. Meyer, is plastic surgeon for the clinics of Lausanne University. The preface is by Prof. Sanvenero-Rosselli of Milan, who is so lavish with his approval and praise that it looks almost as though he was defending the book against adverse criticism in advance. Professor Sanvenero-Posselli prossesses the great attribute of 'ending a ready ear to the opinions of others, however; he is very approachable and since his motives are above suspicion I need not be afraid to state that I largely disagree with his view as far as the figurative representation of the operations is concerned.

In their own introduction, the authors state that they wrote the book in response to the requests of a great many general practitioners and ear, nose and throat specialists. They based it on their

own experiences and on operation methods described in the literature which they considered expedient. They intentionally refrained from illustrating the book with photographic reproductions of results, claiming that to do so would make the book much more expensive and that most authors published only photographs of good results and did not show the course of operations, while the purpose of this book was to inform the beginner on practical problems concerning the situation before and during the operation, without any unnecessary waste of time.

In general, the book is a modern counterpart of Joseph's classic work, which is now a museum piece. This also applies to the character and schematization of the drawings. In as far as it is possible to agree with this technique at all, the drawings are perfect. They are only diagrams, however, and anyone who attempts to work by them is bound to be let down. They present everything in an ideally perfect and simple form — which is never the case in reality. The exact relation of the recommended operation to a clearly characterized defect is likewise not always described.

The book is divided into a general section, which gives a concise review of the history of nasal plastic surgery and discusses the physiology, anatomy and formation of the nose, with more reference to anthropological character than is usually the case. The special section is concerned with corrective and reparative surgery of the nose. In the individual chapters, the authors discuss the indications for different operations and describe the various stages very carefully, often citing a large series of authors, even for the slightest modifications. This complicates the description of the operation itself. It is no doubt a very conscientious method of citation, but in this particular case it seems to be excessive. It comes as a surprise to an old worker to see someone named as the author of an operation which he himself has performed for many years and which was undoubtedly performed by many others before him, because it is quite obvious. It may also happen that some thirty years or so ago he described and illustrated a particular point and now comes across a similar drawing under a name which did not appear in the literature until much later. The literature of Eastern Europe was — and evidently is — not readily available to western authors. Otherwise it must be conceded that the authors have done their best to describe and illustrate the largest possible number of operations, though usually without a critical evaluation of pitfalls and obstacles, especially in the case of those admirable little flaps so perfectly drawn on the ala nasi and the surrounding area. The authors simply content themselves with warning the reader that text-books may create the impression that perfect results are obtained in one operation.

The references are divided according to the chapters. This is very useful to anyone wishing to study the literature, but the amount for the beginner is unnecessarily large.

The authors devote a great deal of space to the correction of bad results occurring after cosmetic operations on the nose and to post-operative complications, among which infection is noticeably frequent. It is surprising to read of colliquation of cartilage grafts necessitating frequent aspiration of an absces. These are obviously cases of infection. A large amount of space is also occupied by periosteal reactions in the form of hyperplasia or osteomas. This is caused mainly by inadequate removal of bone débris after filing and polishing the cut bone surfaces; this should be done very carefully with a curette, at the same time washing out the wound. Immediate reactions are very common, probably because nasal reparative operations are all too often performed under out-patient conditions. The patient ought to be admitted to hospital and remain in bed for

at least 24 hours and ice packs should be applied, so as to limit the formation of haematomas and oedema.

In the chapter on correction of the nose after operation for hare-lip and cleft palate, the authors recommend some operations with which it is impossible to agree and disregard the fact that a large proportion of cases of secondary deformities of the nose after operations for hare-lip and cleft palate are due to the exposure of large areas of bone, including the vomer, the jaw and the palatine bones. Large portions of these bones are left without a covering and undergo secondary epithelization. Extensive sclerosis then develops and contractive scars are formed; these are one of the main causes of the final deformity. As with posttraumatic deformities, emphasis should be placed on preventive care, which can prevent, or at least mitigate, many secondary deformities.

Although the authors publish many methods only from the literature, without evaluating them, the book gives a good picture of nasal plastic surgery and the wealth of western references will be an important source for the specialist working in this field. The publishers are to be congratulated on the way in which the book is turned out. It is a classic example of how a scientific book should be presented. It also demonstrates the growing prestige of plastic surgery — despite its price, the publishers are not afraid that the book will not be sold.

(Akademician F. Burian): Šrobárova 50, Praha 10, Czechoslovakia

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ANNOUNCEMENTS

Announcing the dates of the Tenth Congress of the Pan-Pacific Surgical Association: PART I — September 20—28, 1966 in Honolulu, Hawaii. — Second Mobile Educational Seminar: PART II — September 28 — October 10, 1966 in Japan and Hong Kong. — PART III — September 28 — November 1, 1966 in Japan, Hong Kong, The Philippines, Thailand, India, Singapore, Australia and New Zealand.

The Board of Trustees of the Pan-Pacific Surgical Association is pleased to announce the dates of the Tenth Congress of the Association and the Second Mobile Educational Seminar to countries bordering on the Pacific basin.

Part I, the Honolulu portion of the Congress, will convene at the Princess Kaiulani Hotel in Honolulu, Hawaii, on September 20, 1966 and continue through September 28. Part II and Part III will depart Hawaii on September 28 and travel to Japan and Hong Kong, with Part II returning to San Francisco, California, on October 10 in time for the opening of the American College of Surgeons, and Part III continuing on to the Philippines, Thailand, India, Singapore, Australia and New Zealand, returning to Hawaii on November 1, 1966.

The Tenth Congress offers an extensive scientific program presented by more than 300 leading surgeons from all parts of the world in nine different surgical specialties and related specialties: General Surgery, Ophthalmology, Otolaryngology, Thoracic Cardiovascular Surgery, Neurosurgery, Obstetrics and Gynecology, Orthopedics, Plastic Surgery, Urology, Anesthesiology and Radiology. We extend this invitation to all physicians to attend and participate in these meetings.

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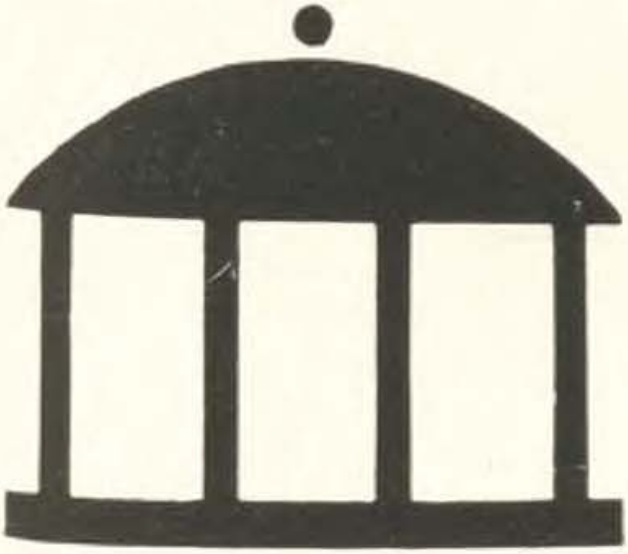

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