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THE MORPHOLOGY OF WOUND HEALING UNDER CRUSTS

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In a previous paper we have described a method for determining the differences in the length of time required by wound healing *(Dévényi* and *Kellner*, 4). The method is based upon measuring the wound area on the first and seventh days, and observing the period of wound healing. Considering that changes in the healing process are due to histological phenomena occurring in the wound, gross examination is unreliable if it lacks a histological foundation. It was, therefore, thought necessary to subject the healing wound to continuous histological examination.

A great number of authors have studied, on various objects, the histology of wound healing in general, and that of healing under crusts in particular *(Marchand*, [11], *Goldzieher* and *Makai*, [8], *Lepeshinskaya*, [10], *Anitchkov*, *Volkova* and *Garsin*, [1], *Huzella*, [9], *Törő*, [15] and [16], *Gaza*, [6] and [7], *Verebély* [17], *Polezhaiev*, [12], *Studitsky*, [13]. Although these authors have described in detail the gross and microscopic changes in their chronological sequence, only few contributions deal with the single histological patterns characteristic of particular, selected points of time *(Anitchkov* and co-workers, [1], *Lepeshinskaya*, [10]). To study that problem was the main purpose of the present experiments.

In order to obtain the characteristic histological patterns presented by the healing wound at certain points of time, wounds were examined in series of 10, at 16 different times. When interpreting the histological findings, special attention was paid to the formation of granulation tissue and of connective tissue fibres, further to the function, structure and change of the crust. We were successful in observing several phenomena the publication of which appears to be a necessary supplement to existing literary data.

Methodology

The experiments were performed on white rats of both sexes, weighing from 130 g to 150 g each. The animals were kept on a mixed diet. The wounds were inflicted by the method previously described *(Dévényi—Kellner*, [4]) and examined after 1, 2, 4, 8, 12, 18, 24, 36, 48 hours and 3, 5, 6, 7, 10, 12 and 14 days, respectively. The animals were killed by bleeding to death. The wounds were excised together with the spinal column, and fixed in neutral formaldehyde, Carnoy's fixing fluid, lead acetate-formaldehyde, and mercury bichloride-alcohol. After embedding in paraffin, serial sections 5 microns thick were made. Staining was made with haematoxylin-eosin, methyl green pyronin, Weigert's resorcin-fuchsin, Heidenhain's iron haematoxylin, toluidin blue (Lyson), Masson's trichrome (modified), Mallory's, van Gieson's, Feulgen's and Giemsa's stains : besides, Gömöri's silver impregnation was also used. Lipoids in the tissues were examined after embedding in gelatin and staining with Sudan III. — A total of 160 animals, in 10 series, were used in the experiments.

Results

Skin excised from the rat's back presents the following layers. (Their enumeration was thought necessary for correct interpretation of the histological pictures.) (i) Epidermis. (ii) Corium, with skin appendages, and, in its lower part, adipose tissue. The quantity of the latter depends on the nutritional state of the animal. (iii) Striated horizontal dermal muscle. (iv) Loose connective tissue with numerous capillaries, groups of fat cells, and a great number of basophil histiocytes around the vessels. (v) Fascia of the back muscle.

The first three of the above layers were removed at the infliction of the wound. The fourth layer, that of loose connective tissue, has thus formed the base of the wound in every case.

1st to 8th hour. The first changes are of a regressive character. The transected skin layers undergo necrosis at the edges of the wound. The necrosis extends in most cases to a marginal hair follicle. The epithelium is shrinking, its cells are becoming pyknotic and stain dark. The collagen fibres of the corium necrotize in narrow strips, the necrosis showing a fibrinous character. Coagulative necrosis can be seen at the site of the transected muscles. A zone of demarcation of varying width, consisting of leukocytes, delimits the necrotized parts. The loose connective tissue at the base of the wound undergoes oedematous swelling. An exudate poor in fibrin appears between the loosened and torn fibres of connnective tissue which are infiltrated chiefly by neutrophil granulocytes. Minute amounts of extravasated blood, leukocytes, a few lymphocytes and macrophages are seen in the neighbourhood of the congested vessels. Scattered in the cytoplasm of the macrophages there are phagocyted red blood corpuscles. Profuse haemorrhage was rare. The wound surface is coated by a thin crust consisting of exudate and a few torn fibres of connective tissue.

8th to 24th hour. The crust is gradually increasing in amount. Apart from dried exudate, it contains shadows of disintegrating cellular and fibrous elements (Fig. 2A). The loosening of the base of the wound continues ; this is, however, no longer simple oedema, since by now a fibrinous network has appeared, with an arrangement parallel to the surface. The filaments of fibrin stain with pyronin first a light, later a bright red, and present a granular-flakelike structure. Mingled with the network of fibrin, remnants of the loosened, torn up original argyrophil fibrous network can be observed (Fig. 3A). A zone of leukocytes separates the secretion from the crust. The place of the leukocytes infiltrating the secretion is taken more and more by macrophages and lymphocytes. Perivascularly basophil histiocytes, together with free granules, originating from their cytoplasm, can frequently be observed.

In the cytoplasm of many macrophages there are vacuoles, mostly next to the nucleus, which they displace to the side. Other macrophages contain a dense

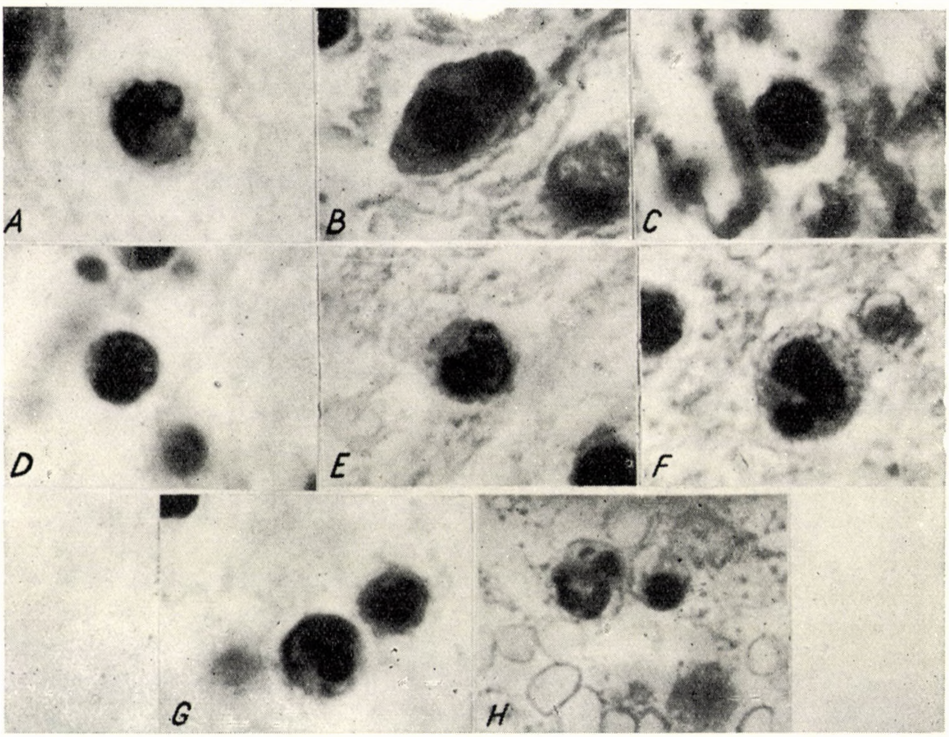

Fig. 1. Formation of »plasmatic nucleus«. A—C: Densification of diffuse nuclear substance in the cytoplasm; D—G: Globules, intensely staining with nucleus stains, in the cytoplasm; H: Extracellular globule surrounded by faint protoplasm (methyl green-pyronin, $\times 1600$)

area, staining dark red with methyl green pyronin, and merging into the surrounding cytoplasm without a distinct margin. In other cells this dense part stains purplish or blue, assuming a spherical shape and standing out distinctly of the cytoplasm. It is surrounded by a fine, pale halo. Developed to this stage, the spherule, which we have termed »plasmatic nucleus«, stains intensely with nuclear dyes. The original nucleus of the cell is more and more displaced to the side, embracing in the form of a crescent the fully developed »plasmatic nucleus«. The spherule, moving gradually towards the edge of the cell, becomes, at last, liberated from it. After this, a small area in the mother cell, presumably that

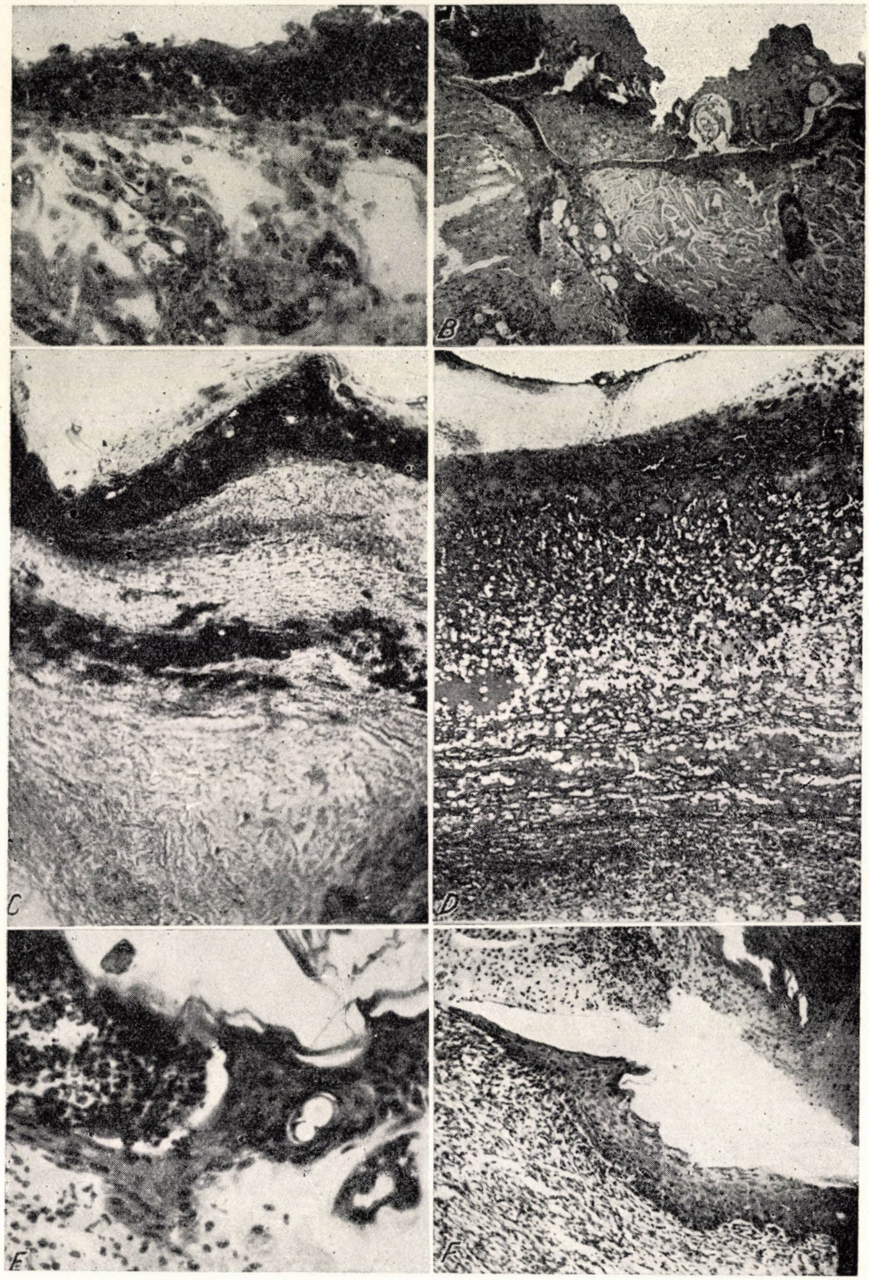

Fig. 2. A : Wound after 12 hours. Surface covered by thin crust. (Trichrome, ×370) ; B : Wound after 48 hours. Parts of necrotized corium in the crust. Beneath it, forward-creeping epithelial tongue. (Haematoxylin-eosin, ×100) ; C : Wound after 36 hours. Parts of adipose tissue in the crust. (Sudan III, ×100) ; D : Wound after 5 days. Characteristically stratified crust. (Trichrome, ×100) ; E : Wound after 1 day. Bifurcating epithelium. (Haematoxylin-eosin, ×370) ; F : Wound after 6 days. Keratinization of forward-growing epithelium ; detachment of crust. (Haematoxylin-eosin, ×100)

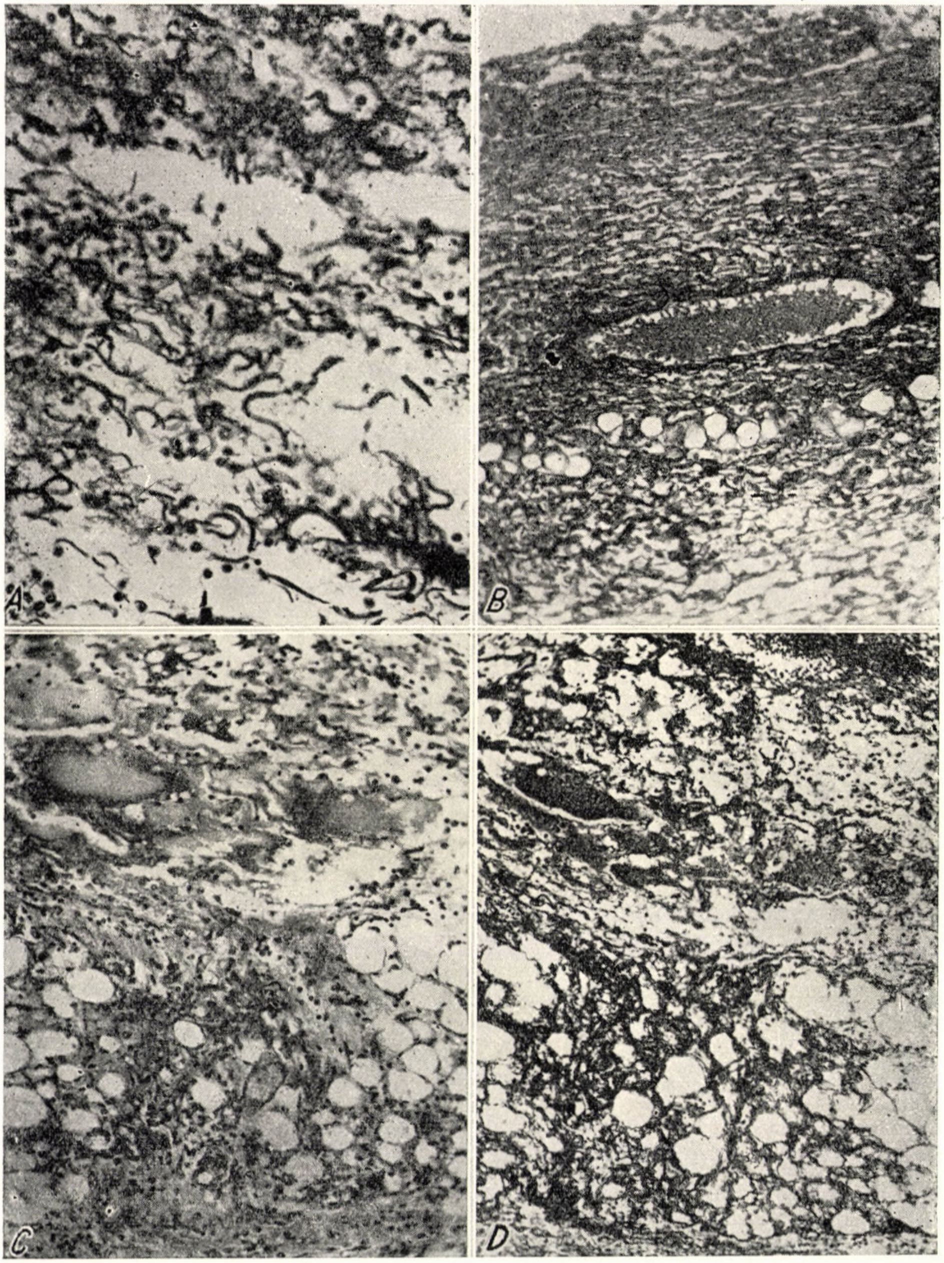

Fig. 3. A : Wound after 1 day. Disintegration fibres and granularly impregnating fibrin in the wound exudate. (Gömöri's silver impregnation, ×370) ; B : Wound after 2 days. Fine connective tissue fibres and fibrin network ; negative to fibre stains. (Mallory, ×180) ; C : Wound after 2 days. Perivascular focus of granulation tissue. (Toluidine blue, ×180) ; D: The same with Gömöri's silver impregnation

from which the spherule originated, stains a brighter red than the rest of the cytoplasm. The nucleus of the mother cell retains its original structure from first to last. While some of the extracellular spherules are naked, others are surrounded by a fine, pale, elliptical strip of cytoplasm. We were not able to follow their further evolution (Fig. 1. A—H).

24th to 36th hour. The wound is covered by a thick crust that stands out of the surface of the skin. We find in its substance not only the above described elements but the necrotic and demarcated parts of the wound-edge as well (Fig. 2B). The richly vascularized layer of adipose tissue bulges out of the secretion in varying degrees, attaining in some areas the lower edge of the crust. In preparations stained with Sudan III, isolated group of fat cells may be observed in the crust (Fig 2C).

The epithelium thickens in the form of knobs on the edges of the wound. A proliferating, many-layered epithelial tongue has grown out of it, extending partly to the surface of the crust and partly underneath it (Fig. 2E).

The epithelial tongue under the crust rests on a base made up of leucocytes and a mass of fibrin, and frequently terminates in a broomlike ramification. The epithelial cells are elongated, they stain faintly, their intercellular substance is indistinct, the nucleolus is small or missing ; they resemble cells of connective tissue. No keratinization of keratohyaline granules can be seen as yet, nor are mitotic forms observable. The epithelial tongue above the crust is often loose, infiltrated by leukocytes. In some places there are isolated groups of epithelial cells surrounded by leukocytes. The epithelium, grown onto the surface of the crust, always presents keratinization, its cells are markedly pyknotic.

48th hour. Apart from infiltration by macrophages, isolated foci of proliferating fibroblasts and newly formed capillaries can be seen around the vessels of the abundantly vascularized layer of adipose tissue (Fig. 3C). Impregnated preparations reveal in these areas also newly formed argyrophil fibres, connected with the vessels and the adipose tissue (Fig. 3D). There are only traces of intercellular ground substance showing metachromasia on staining with toluidin blue. The loosely structured exudate impregnates with silver. Argyrophilia is bound in the first place to the remnants of the disintegrating original argyrophil fibre system (Fig. 3A). The horizontally arranged network of fibrin impregnates in granules. Staining according to Mallory and van Gieson revealed that the network although it impregnates with silver, does not possess the properties of fibres (Fig. 3B).

Third day. Foci of granulation tissue appear, situated in chains along the amply vascularized layer of adipose tissue. There is an abundance of intensely metachromatic intercellular ground substance in these foci. The number of argyrophil fibres has also increased. Newly formed granulation tissue is seen in both edges of the wound ; its fibres show a vertical course (Fig. 5A). Being a direct continuation of the loose submuscular connective tissue, it is in no way connected with the more centrally placed focal granulation tissue. The surface of the marginal granulation tissue is covered by epithelium in which signs of differentiation can be observed. The basal layer has developed by this time, and keratohyaline granules, too, are found in some sections. The crust itself has an unchanged aspect as compared with the preceding phase.

Fifth day. The wound cavity is completely filled by granulation tissue. The granulation tissue arranged in foci in the centre cannot be distinguished from the vertically arranged marginal granulation tissue. The fibres are lying horizontally throughout the wound, and the intensely metachromatic intercellular ground substance is present everywhere.

On the edges of the wound the surface of the granulation tissue is covered by epithelium of varying width. The epithelial tongue is elongated, taking a wedgelike shape, and shows definite keratinization in its thicker, marginal parts. There is no clear-cut borderline between granulation tissue and crust in the areas not covered by epithelium. Serous imbibition, further an infiltration composed of monocytes and leukocytes, and also disintegrating fibres, are found in the upper layer of the granulation tissue, while in the lower part of the crust next to the granulation tissue there are necrotic, but in their contours still recognizable, rests of granulation tissue and remnants of fibres.

The entire surface of the wound is covered by the crust. While adhering closely to the granulation tissue in the centre of the wound, on the edges above the forward-creeping and hornified epithelial tongue it shows a loose structure.

The crust consists of characteristic layers (Fig. 2D). On its surface there is a thin, homogeneous, desiccated zone. Below this, a broad homogeneous substance is seen, with the debris of vessels and fibres, and some groups of fat cells ; this is followed by a leukocyte layer, and then by detached fragments of cells and fibres embedded in a homogeneous ground substance. The lowest layer of the crust consists of loose connective tissue imbibed with serum, its interstices including red blood corpuscles and macrophages. This layer passes imperceptibly into the intact, newly formed granulation tissue. At the time when the crust tonches the exudate no more, but has come into contact with granulation tissue, it is the nocrobiotic, necrotic granulation tissue, and no longer the leukocytic zone, which forms the lowest layer.

Sixth day. In some of the sections the surface is covered by the above described original crust. The hornification of the epithelium causes, just as on the fifth day, its edges to detach themselves from the base (Fig. 2F), whereas, in other sections, no crust of this type occurs. In the central parts uncovered by epithelium, a thin, newly formed crust containing destroyed fibres, vessels and cell-debris appears, produced by the necrosis of the uppermost parts of the granulation tissue.

It was observed in previous experiments *(Dévényi—Kellner*, [4]) that, in the course of the healing of wounds inflicted in rats, a spontaneous change of crust took place between the 5th and 7th day. This phenomenon might explain the above described two types of histological pattern noticed on the 6th day.

With a view to clarifying this process, the crust was removed shortly before the 6th day, the anticipated time of the change of crust. Sections, made one half hour after removal of the crust, revealed necrosis in the superficial layers of the granulation tissue (Fig. 4A). By the following day, a new crust, containing cell and fibre debris, has developed (Fig. 4B). The necrosis of superficial layers was restricted to the surface of granulation tissue not yet covered by epithelium.

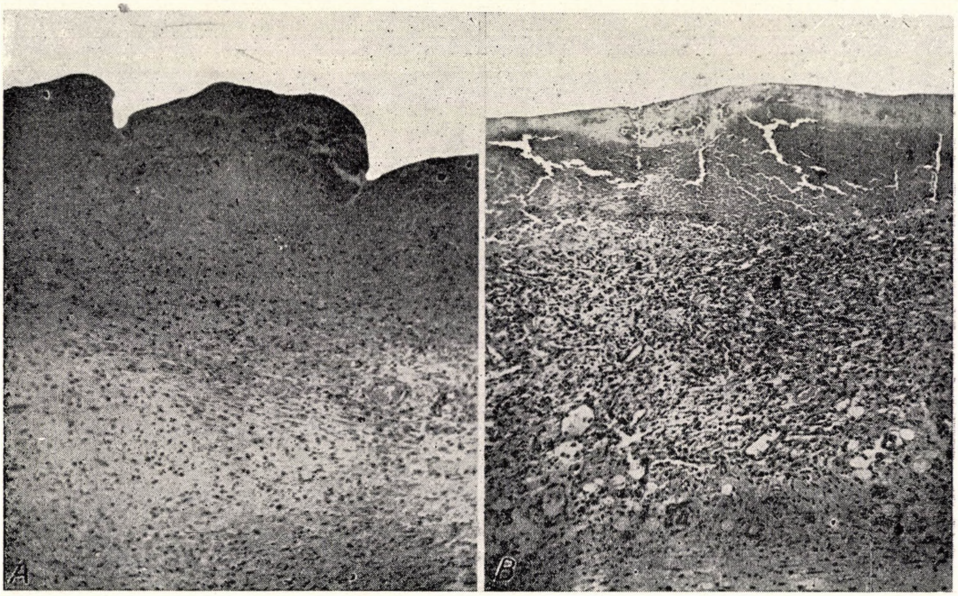

Fig. 4. Wound surface after removal of crust. A : After half an hour ; B : After 1 day ; wounds after 6 days. (Haematoxylin-eosin, ×100)

Irrespective of the condition of the crust, the entire wound cavity appears to be filled with granulation tissue on the 6th day. The fibres of the granulation tissue are arranged parallel to the surface (Fig. 5B). The vascular structure has no definite pattern. Everywhere a great amount of intensely metachromatic ground substance occurs. Isolated fat cell groups are found in the granulation tissue, chiefly in the lower layers.

7th day. The granulation tissue consists of the following well distinguishable layers (Fig. 5C), from up to down,

(i) A layer of loose fibre with vertically running vessels. The fibres are stretched out between the vessels and run parallel to the surface.

(ii) A layer of horizontally arranged coarse fibrous structure with decreased argyrophilia and vessels with varying courses.

(iii) A layer of collageneous fibres with horizontally running vessels and scattered groups of minute fat cells.

(iv) A layer of normal loose suprafascial connective tissue.

Metachromatic ground substance is found in the two upper layers only, its quantity being smaller than on the 6th day.

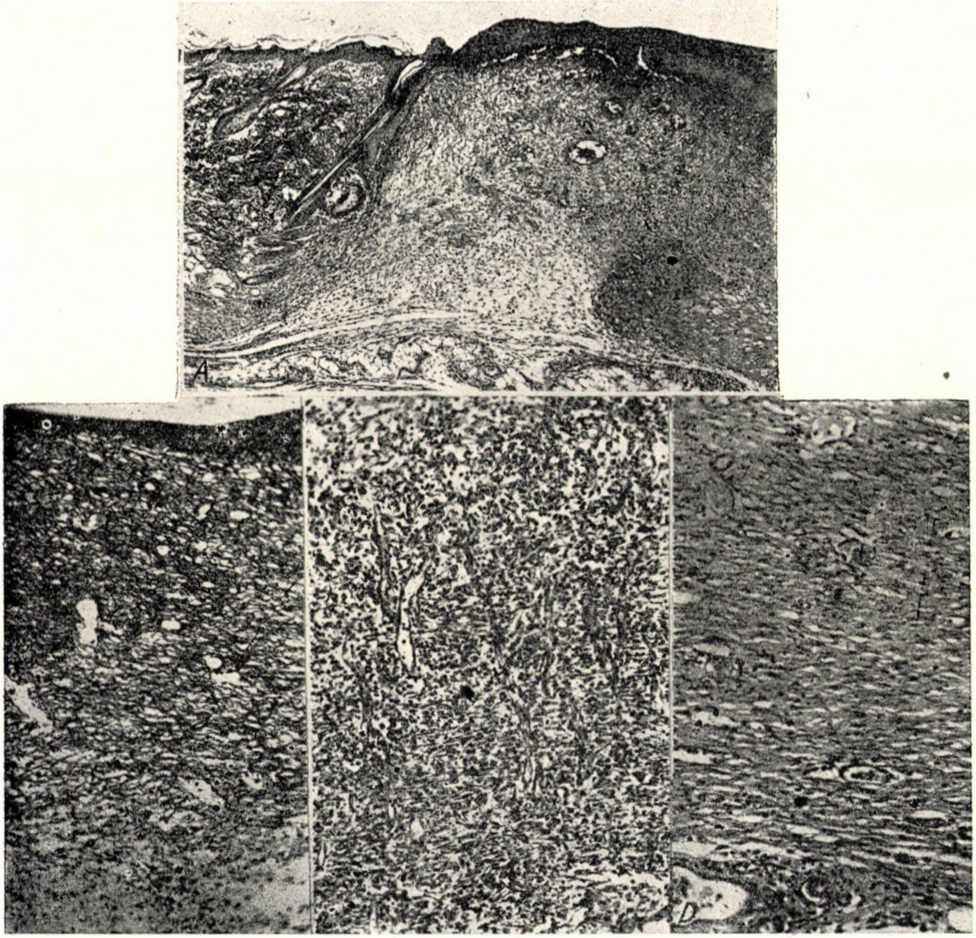

Fig. 5. A : Wound after 3 days. Vertically arranged granulation tissue in the corner of the wound cavity. (Trichrome, ×49) ; B : Wound after 6 days. Horizontal argyrophil fibrous network. (Gömöri's silver impregnation, ×180) ; C : Wound after 7 days. Granulation tissue with vertically running vessels. (Toluidin blue, ×180) ; D : Wound after 12 days. Collagenous fibres. (Haematoxylin-eosin, ×180)

The majority of the cases present alterations characteristic of the change of crust that has taken place on the surface. The thin crust is composed of necrotized granulation tissue, and extends to the unepithelized areas only. The epithelial tongue, grown forward, adheres in its whole extent to the mature granulation tissue. The surface is covered by horny epithelium, and in the upper layers keratohyaline granules occur.

10th, 12th and 14th days. The layers of the granulation tissue become increasingly indistinct (Fig. 5D). The amount of cellular substance and the richness of vascularization decrease, the walls of the remaining vessels become thicker or, in some places, undergo hyaline degeneration ; the fibrous structure becomes denser, and gradually assumes a collagenous character. The granulation tissue of the wound cavity can be sharply distinguished from the corium even on the 14th day ; the accessory organs of the skin cannot be traced in it.

In part of the wounds examined, their entire surface was covered by epithelium on the 14th day. Keratinization causes the crust to become loose and, then, to detach itself. The borderline between the epithelium and the connective tissue of the epithelized wound is straight ; it has no papillary structure. In another part of the wounds, complete epithelization had not yet developed by the 14th day.

Discussion

Morphological changes, characteristic of particular points of time

Methods are available which, with the aid of a mathematical formula, allow to infer the degree of healing from the size of the wounded area and the date of healing. *(Carrel* and *Nouy* 2 ; *Dévényi* and *Kellner* 4). All of these methods lack, however, a histological background.

Histology revealed that characteristic morphological patterns are bound to certain definite points of time.

(i) On the 2nd day of the healing process, there are perivascularly arranged foci of fresh granulation tissue at the base of the wound, containing some metachromatic intercellular ground substance and a thin argyrophil network.

(ii) On the 7th day, the granulation tissue shows a number of well distinguishable layers, in the uppermost of which a transversal, loose network of fibres can be between the vertically running vessels.

We consider these two histological pictures characteristic of the 2nd, respectively the 7th day since in the series examined they occurred regularly on those days. These two morphological data, in conjunction with the previously described planimetric method, are, in our opinion, suitable for being employed as a test for determining the degree of healing. This combined test appears to be well suited for the serial examination of substances which promote healing, because it allows not only of gross observation but offers also the possibility of examining the histological occurrences, and further, because it requires a limited number of animals for the histological examination.

We want to make it distinctly understood that the method has been applied in this form to rats only, and with just a chosen size of wound. It is well conceivable that a morphologically different picture would be obtained if we were to apply it to the healing process in some other animal species. This possibility seems to be confirmed by the investigations of *Anitchkov* et al. [1] who, when observing wound healing in cattle, rabbits and humans, found the mechanism of healing to depend on the structure of the skin.

Different ways of karyogenesis in the exudate of the wound

It is generally believed that part of the cellular elements of the wound-exudate originates from the blood, while another part of it is being supplied by the mobilized cells of the neighbouring connective tissue which wander into the wound cavity.

Lepeshinskaya [10] traces karyogenesis back to the granules produced by the decomposition of blood. She thinks the granules are phagocyted by migrating cells ; these, in their turn, change into basophil histiocytes, which, when disintegrating, give rise to living granules. She observed every possible transitory form from tiny granules to lymphocytes.

Observations made by *Törő* [16] showed that migrating cells, phagocyting red blood corpuscles, were transformed into histiocytes. On the other hand, he did not see lymphocytes to be formed from granules produced after the disintegration of histiocytes. According to Törő, extensive klasmatosis occurs in tissue cultures, resulting in the genesis of minute Feulgen-positive globules of cytoplasm which contain nuclear substance. It could not yet been ascertained whether they are detached cell fragments or forms of cell-growth. They contain an ample amount of ribonucleic acid, and this seems to point rather to a synthetic process.

The conclusion to be drawn from these investigations is that, while some of the cells in the secretion may have migrated into the healing wound, others must have developed locally.

Examining wounds in an early phase of the healing process, we observed a different form of karyogenesis. We saw the diffuse nuclear substance, as contained in the plasma of vacuolated macrophages, to condense with increasing pyroninophilia ; this process of condensation produces a kind of spherically shaped nuclear matter, i. e. the so-called »plasmatic nucleus«, which is highly sensitive to nuclear stains; detaching itself from the rest of the cytosplasm, it finally becomes liberated. The free globule is frequently surrounded by protoplasm. Although we had no opportunity to observe the subsequent development of the nuclear structure in these globules, we assume that they develop into genuine cell.

This process of cell birth is, on the whole, in agreement with the neokaryogenesis observed in thymus cultures by *Törő* [15]. Histological methods in themselves are, however, insufficient for definite conclusions to be drawn in this respect.

We did not encounter either profuse haemorrhage or phagocytosis in the material. On the other hand, we were able to observe cellular infiltration and basophil histiocytes in every case. The basophil histiocytes, these well known cells of the rat's skin *(Sylvén*, [14],) frequently disintegrated into granules. We could observe the transformation of macrophages into basophil histiocytes, or the development of new cells from the granules by the disintegration of the basophil histiocytes.

Fibrillogenesis and the formation of granulation tissue

According to *Huzella* [9], mere physical factors may cause fibrous structures to arise from the wound-exudate, but, on the other hand, they may be produced also by the immigrating cells investing filaments of fibrin with a secretion which, after solidification, changes them into connective tissue fibres. *Törő* [16] assumes that fibres may arise on the surface of healing wounds, as it is here that physical forces come into full display and fibroblasts are the last to appear. According to *Lepeshinskaya* [10] and *Törő* [16], it is possible for fibres to develop from blood and exudate, both of these consisting of living matter. *Lepeshinskaya* observed granular precollagen in wounds of 3 hours, and homogeneous, collageneous fibres in those of 5 hours. She presumes that fibrin, which first exists in the sol form, becomes a gel, to turn then into precollagen, and subsequently, after intensive hydration, into collagen. *Sylvén* [14] considers the intercellular metachromatic substance, appearing simultaneously with fibroblast proliferation, to represent the ground substance of the fibrous system. As mentioned before, *Anitchkov* et al. [1] hold fatty tissue to be the source of granulation tissue. According to them, the fatty tissue disappears in proportion to the growth of granulation tissue; the wound cavity is filled gradually by the upward proliferating young granulation tissue. The upper parts become necrotized, while in the deeper layers gradual symptoms of maturation can be observed.

Aware of the above data, we paid attention to fibrillogenesis in the course of healing. We examined, first of all, whether a direct transformation of fibrin into fibres takes place. It was found that in a very early phase (within 48 hours), when the connective tissue has not yet begun to proliferate, the fibrin network, displaying a horizontally arranged fibre-like structure, impregnates with silver. It was, however, also found that argyrophilia is mostly confined to destroyed fibres left over from the original ground tissue, found in the interstices of the fibrin network, whereas fibrin itself impregnates granularly. By comparing our specimens impregnated with silver and those stained with other dyes (Mallory, Van Gieson), it becomes clear that in the secretion only disintegrating fibres stain adequately (blue and reddish-yellow, respectively), whereas the fibrin network stains a bright red with Mallory's dye and yellow with Van Gieson's stain. In later phases, regular fibre structures occur only in proliferating granulation tissue. We are of the opinion that the granular argyrophilia of the fibrin-network does in no way indicate the change of fibrin into fibre.

Our investigations have shown the richly vascularized connective tissue, containing fat cells and found at the base of the wound, to be the source of the granulation tissue with which the cavity of the wound is filled. This layer undergoes oedematous loosening at an early stage, and arches out into the wound cavity. Its proliferation begins in perivascular foci. Development of fibres was found to be bound to these foci of granulation tissue, and the number of fibres to increase in proportion to the quantity of granulation tissue. The process starts at the base of the wound ; filling the wound cavity by degrees, it pushes the secretion upwards, which latter, together with the uppermost layer of granulation tissue, loses itself in the crust in the course of healing. Simultaneously with the first signs of proliferation, the metachromatic ground substance appears. This substance increases, together with the quantity of argyrophil fibres, in proportion to the increasing proliferation, and disappears during the period of collagenization.

Significance of the crust. Spontaneous change of crust

Protecting the wound against outside irritation, the crust may control the growth of epithelium and serve as a culture medium for proliferating tissues. (*Törő* [16], *Anitchkov* et al. [1]. This mutual relationship was formerly held to have a biological significance, assuming that growth-promoting substances arise from the deteriorating tissues. (Wound hormone, necrohormone, trephone, desmone.) Examining these materials, *Davidson* [3] emphasized that, though their existence has never been definitely proved and none of them has been chemically characterized, one may still conclude to their existence. *Lepeshinskaya* [10] has thrown an entirely new light on the biological significance of deteriorating tissue, stating that the destruction of cells involves the genesis of living matter, living protein, which, independently of the cells, may give rise to new structures.

The spontaneous change of crust, systematically observed by us and in respect of which we have found no data in the literature, has been proved by our investigations to be a growth promoting factor. The original, thick, rigid crust fixes the edges of the wound and keeps them apart. It is probable that, from a certain point of time onward, it presents no longer a suitable milieu either to the epithelium or the connective tissue. After the detachment of this crust the wound contracts, and a new crust, considerably smaller than the original, is formed on the unepithelized wound surface. It is made up of necrotized granulation tissue.

Epithelium grows rapidly under the fresh crust. The immediate cause of the spontaneous change of crust is the keratinization of the marginal epithelium. We trace the dwindling of the wound back to the shrinkage and the accompanying exsiccation after the change of crust, though it is conceivable that a contraction of the fibre network in the granulation tissue is also as a factor in the process.

Epithelization

According to the literature, regeneration of the epithelium begins immediately after the wound has been inflicted (*Anitchkov* and co-workers [1], *Dunn, Glückman* and *Tansley* [5]. *Törő* [16]) observed the epithelium to creep forward with a peculiar, collective movement. No mitoses have been found in the forward-creeping epithelium. In connection with the early epithelial proliferation, a certain importance must be attached to the fact that the necrosis of the wound edges extends, as a rule, to a hair follicle. There exists a relationship between the hair follicle and the knobby thickening of the marginal epithelium from which the epithelial tongues grow out like feelers. It may be assumed that, apart from the superficial epithelium, epithelial cells of the hair follicle may also play some part as a source of proliferating epithelium. Our own observations were in agreement with those of *Anitchkov* et al. [1] in that the epithelium grows temporarily on to every possible surface. This seems to be confirmed by the fact that, in the early stage of the healing process, the epithelium spreads both over and under the crust. Such epithelial tongues perish, however, in every case. Their adhesion does not become durable unless their base consists of mature granulation tissue. Fundamentally, the same holds true in respect of the granulation tissue, the superficial layers of which are destroyed as long as they remain uncovered by epithelium.

It can be seen from the above that the healing of wounds results from a dynamic interaction of granulation tissue, epithelium and crust.

Summary

The morphological changes occurring under the crust during wound healing have been examined in 160 white rats. On the second day after infliction of the wound, circumscribed foci of granulation tissue were found at the base of the wound ; on the seventh day, regular layers of granulation tissue were observed, and the uppermost layer contained vertically running vessels. We consider these histological pictures to be characteristic of these two days.

Together with our quantitative test, this histological test is, in our opinion, suitable for the determination of the degree of healing.

We observed the phenomenon of a change of crust to take place in the course of the healing process ; this happens on the 5th to 7th day, as a rule. After having cast off the crust, the wound becomes smaller, and a new crust is formed, chiefly from the necrotized superficial parts of the granulation tissue.

The phenomenon of nuclear genesis was observed on an early phase of the healing process. Densified, the diffuse nuclear substance of the cytoplasm forms a so-called »plasmatic nucleus«

which becomes liberated from the cell. A faint rim of cytoplasm is formed around the extracellular body.

According to our findings, the development of connective tissue fibres is bound to foci of granulation tissue.

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НОВЫЕ ДАННЫЕ К МОРФОЛОГИИ ЗАЖИВЛЕНИЯ РАН ПОД КОРКОЙ

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Резюме

Мы исследовали заживление кожных ран у 160 белых крыс., в 16-и стадиях, начиная от одного часа после нанесения раны до 14 дней. Характерная морфологическая картина наблюдалась на второй и на седьмой день. Мы установили, что на дне раны образование грануляционной ткани начинается на второй день в виде периваскулярных очагов. Эти отдельные очаги расширяются и в конце концов, — сливаясь, — выполняют раневую полость. В седьмой день в грануляционной ткани отмечается закономерная слоистость, характерными являются вертикальные сосуды, расположенные в верхнем слое. Описанные гистологические картины являются характерными для упомянутых дней, и мы считаем эти картины — на ряду с описанной нами-же пробой — надежным гистологическим показателем степени заживания.

Во время заживания поверхность раны покрыта коркой. В 5—7 дней вследствие ороговения продвигающегося эпителия связь корки с дном раны разрыхляется. После этого рана уменьшается в размере и на поверхности еще не покрытой эпителием, образуется новая корка. Эта корка состоит из поверхностной некротизированной части грануляционной ткани.

В 12—36 часах мы наблюдали в раневом секрете явление образования новых ядер. В макрофагах разлитое ядерное вещество протоплазмы собирается в ограниченном участке и образует т. н. »плазматическое ядро«. Последнее становится шарообразным и выбрасывается из клеточного тела. Вокруг выброшенного таким путем образования возникает светлая кайма из протоплазмы.

Образование соединительно-тканных волокон происходит исключительно в очагах грануляционной ткани и количество этих волокон возрастет параллельно с грануляционной тканью.