CHANGES IN THE CUTANEOUS NERVES IN LEPROSY [1]

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It has long been suspected that leprosy is primarily neural in its inception. The peripheral nerves in this disease have been studied by many workers in the past. According to Klingmüller (10), Danielssen and Boeck were the first to describe the changes in them. The famous Dehio-Gerlach theory (17), that the finest cutaneous nerve twigs were the first to be affected by the disease process, which then crept up centripetally, was first propounded toward the end of the last century. Since then many reports have appeared in the literature dealing with the pathological anatomy of the peripheral nerves in leprosy. Although many of the fundamental principles of this aspect of the disease have been enunciated, interest in the subject has been renewed with increase in the knowledge of the nerve supply of the skin and of the histology of the early lesions of leprosy.

The present work is concerned with a histological study of the nerves in skin biopsies obtained from different types of lesions of leprosy. An attempt is made to correlate the clinical and histological findings in these cases.

MATERIALS AND METHODS

Thirty-two pieces of skin were obtained from 30 leprosy cases showing typical lesions. Of these specimens, 19 were from lepromatous cases, 8 from tuberculoid lesions, 3 from maculoanesthetic lesions, and 2 from prelepromatous ("juvenile" or "incipient leproma") lesions. In the first three groups, cases in different stages of evolution as well as resolved lesions were included. Most of the specimens were obtained from the back; a few were from the forearm and chest. The tissues were fixed in 10 per cent neutral formalin, and frozen sections 20 to 30 microns thick were cut and stained by a modified Bielschowsky silver impregnation method for nerve fibers. On an average six sections were studied from each specimen. Paraffin sections were also made, and routinely stained with Ehrlich's hematoxylin and eosin; and for acid-fast bacilli by a modified Fite method.

OBSERVATIONS ON THE ALTERATION OF CUTANEOUS SENSIBILITY

Our observations were limited to tacticle sensibility only, which was tested for with the tip of a fine feather. Four types of stimuli were

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applied: a light touch at one spot at a time; a light stroke over the lesion; a heavier touch so as to bend slightly the hairs of the feather, but still limited to a spot; and, fourthly, a heavier stroke over the lesion. The lesions were carefully examined with the aid of a hand lens, and wherever hair was found, and wherever the sensation of touch had not completely disappeared, the area was shaved closely, and the tests repeated. The contralateral site was always examined in the same way. Thirty-five cases of different types of leprosy were examined in this manner, and of these 25 were biopsied.

It was found that the normal skin on the contralateral site which responded to light touch often failed to respond to the same after shaving. In the maculoanesthetic lesions and tuberculoid lesions any persistence of response to light touch was always associated with the presence of hairs, although the reverse was not true. After shaving, the lesions were either completely anesthetic to all the four types of stimuli, or responded occasionally to a heavier stroke, which probably stimulated the base of the hair shaft, more by pressure than by touch.

In one of the cases of incipient leproma, the macules were completely anesthetic to touch after shaving. In the early lepromatous cases, hairs were always present, although of a fine, downy type. The skin in these cases responded to light touch and stroke, but the response to stroke after shaving was variable. However, the consistent alteration in the response to the different grades of stimuli was quite unexpected. In the advanced lepromatous cases the skin had often lost all the hair and was completely anesthetic, or where hair persisted complete anesthesia was noticed after shaving.

CHANGES IN THE NERVES

The changes in the nerves as seen in the routine sections stained with hematoxylin and eosin, and for acid-fast bacilli, were the same as have been described in the literature. Early maculoanesthetic lesions showed an inflammatory infiltrate in foci which often took the form of branching strands corresponding to the branching fibers of the cutaneous nerves, an observation that has been recorded by Dehio (4) and by Klingmüller (10). Some of the changes observed in silver-impregnated sections regarding the Schwann cells, and the infiltrate, were also observed in the routine-stained sections. In silver-impregnated sections the epithelium in practically every case showed numerous argentophilic granules in the basal layers, so that it was not possible to study the course of nerve twigs after they entered the epithelium.

(1) In maculoanesthetic lesions the immediate subepithelial part of the corium showed either a complete absence of nerve fibers or only an occasional fiber imbedded in the infiltrate. In the mid corium there were variable numbers of fibers, often inside the focal infiltrate, either singly or in narrow bundles. In the deeper layers of the corium several nerve bundles could be identified. These showed a less compact arrangement of

fibers than is seen in a normal nerve, and occasionally there was a complete absence of fibers. In such instances only a wavy outline of the bundle could be seen. The fibers when present were separated from each other, giving a splayed appearance (Fig. 1). Apart from the collection of inflammatory cells around the nerve bundle, cells were also seen inside the bundle separating the fibers. The latter often showed an irregular contour of their surface, and were tortuous. Alongside thicker fibers, very thin ones were also seen. The hair follicles were surrounded by a varying amount of infiltrate and showed either no nerve fibers or an occasional fine fiber ending in them.

(2) In tuberculoid lesions the subepithelial part of the corium showed practically no nerve fibers, or an occasional fine one in the infiltrate. The middle and deeper parts of the corium showed only occasional solitary fibers and bundles imbedded in inflammatory infiltrate. The number of fibers encountered in this type of lesion was much less than in maculoanesthetic lesions. The bundles in the deeper layers showed much splaying of the fibers inside them, the space between the fibers being occupied by inflammatory cells and Schwann cells (Fig. 2). The fibers in the diverging sheaf showed much variation in their thickness, some being very thin and some showing fusiform bead-like swellings along their course (Fig. 3). Fragmentation of the fibers was also frequent (Fig. 4). Old lesions and resolved lesions showed a complete absence of nerve elements, or only short fragments of fibers. The findings in hair follicles also varied with the age of the lesions, the older ones showing a complete absence of fibers. When nerve fibers were present, their number varied in the different hair follicles.

(3) In the "incipient" lesions (prelepromatous variety) the subepithelial corium showed a few fine fibers running in different directions and entering the epithelium. Some of them were in the inflammatory infiltrate. In the middle part of the corium a few solitary fibers were found, but only in the infiltrate. These usually were thin and with only slight argentophilic properties, or they showed irregular thickenings along their course, giving a beaded appearance. Fragmentation was also seen, the fibers breaking up into wavy and twisted segments. The deeper corium showed several bundles of fibers with some separation and proliferation of Schwann cells. The hair follicles were well preserved, and many of them were free from infiltrate and showed a number of fine fibers ending in them.

(4) In the early lepromatous cases a variable number of fibers was found in the subepithelial corium. Some of them were horizontally placed in the "clear zone," and others coursed vertically and obliquely through the infiltrate, crossing the "clear zone" and entering the epithelium (Fig. 5). In advanced cases, practically no fibers could be found in this part of the corium. In the mid corium, in the band of infiltrate, a moderate number of fibers—single or in narrow bundles—could be seen coursing

through the infiltrate in a wavy fashion. The fibers were more in number than were encountered in other types of lesions, and it was very common to find apparently normal fibers which could be traced over a long distance in the infiltrate (Fig. 6). In the advanced cases the number of fibers found in this part of the corium was far less than in others, and much fragmentation was evident (Fig. 7).

The fibers in all cases showed fusiform swellings along their course, giving a knotted or beaded appearance (Fig. 8), and thinning of the axons, much varicosity, and fragmentation. The fibers often appeared flattened and, instead of being cylindrical, looked like fine twisted cotton fibers. Some of the small bundles showed a proliferation of cells with spindle-shaped nuclei. Another frequent finding was the occurrence of saccules or bubbles along the course of the fibers. These were apparently inside the axon, since their walls showed the same argentophilic property as the main fibers. In transversely-cut nerve bundles, thick fragments of fibers and much thickening of the perineurium was encountered (Fig. 9).

In the deeper corium, as a rule, many bundles of nerves were found except in advanced cases; in them none were seen, or only fragments or shadowy outlines of bundles were present. The bundles when present were free, or surrounded by infiltrate, but usually they showed fewer fibers than normal, those present being separated from each other by Schwann nuclei (Figs. 10 & 11). The separation of fibers was, however, much less apparent than that seen in tuberculoid lesions, and the divergence of fibers from each other was less evident. Occasionally, it was possible to find an apparently normal bundle becoming progressively involved in the infiltrate, the fibers becoming less and less in number as the cellularity increased, and finally getting lost in the cell mass. Except in very advanced lepromatous cases, the hair follicles were fairly well preserved. These were partly or wholly surrounded by infiltrate and with a moderate number of fibers ending in them (Fig. 12). These fibers often coursed through an area of inflammatory infiltrate and entered the follicles. Occasionally there were encountered hair follicles apparently free from infiltrate but without any nerve fibers.

DISCUSSION

We have limited our observations to the changes in the free nerve endings serving tactile sensibility. The changes in the Meissner's corpuscles and Pacini corpuscles have been described by Hoggan (8), Ssudakewitsch (13) and, more recently, by Alvarez *et al.* (1). The frequent involvement of the nerves in all of the skin lesions has been described by Grieco (6). Alvarez and associates, using gold and silver impregnation methods, have confirmed the old concept that the leprous process shows an affinity for the small nerves of the cutis and subcutis. The differences between the changes in nerves in lepromatous and tuberculoid lesions

have been studied by many, although most of these reports were concerned with advanced lesions. Bernucci (2) stated that in nodular leprosy nerves were unchanged and that not even degeneration was observed. But Hashimoto (7) found degeneration of free nerve endings, both in "leprous granuloma" and in "macular" lesions.[2] Samgin (12) was of the opinion that the changes in nerves of maculoanesthetic lesions, apart from the difference in the number of bacilli, were the same as found in lepromatous lesions. More recently Decoud (3) has expressed the view that nerve injury is a constant feature in both types. According to him the fibers persist for a long time in lepromatous cases, while in tuberculoid lesions they are destroyed early.

Our findings are in general agreement with the ideas of Decoud and others, that nerve fibers are damaged in both types of the disease. The involvement and destruction of the subepithelial free nerve endings was a fairly constant feature in all types of lesions, except in early cases of lepromatous leprosy. On the other hand, in the maculoanesthetic and tuberculoid lesions the free terminals appeared to have been destroyed very early. In the lepromatous skin they persisted for a long time and could often be traced from the depth through the band of infiltrate to the clear zone below the epithelium. In advanced lepromas, however, the free terminals disappear completely, which would account for the absence of tactile sensation in these cases.

The finding of nerve twigs imbedded in the infiltrate, and the fragmentation and beading of axons, have been described by previous workers. The absence of nerve fibers in the foci of infiltrate may either mean that they have been completely destroyed, or have lost their argentophilic property. The latter possibility has to be kept in mind, since we have frequently found very lightly stained fibers side by side with darker ones. The proliferation of Schwann cells in the nerve bundles has been considered by Alvarez *et al.* (1) and by Fite (5) as nonspecific, and attributable to Wallerian degeneration. We have gained the impression that the separation of nerve fibers inside the bundle in tuberculoid lesions is due to the infiltration by inflammatory cells, which appear to choke the fibers to death; while in lepromatous lesions the separation is produced by proliferating Schwann cells replacing the degenerated nerve fibers. But too much stress cannot be laid on this point since, as Fite (5) has observed, the identification of Schwann cells in an infiltrated nerve is a doubtful proposition.

The examination of nerve endings in hair follicles has been very interesting. Stein (14) considered that the follicular apparatus was involved in 33 per cent of cases and that this structure formed the starting point of leprous granulomata. We have found that the destruction of hair follicles depends on two factors: the duration of the lesion, and the depth to which infiltration occurs. While in tuberculoid cases the fibers

[2] In Japanese terminology, the word "macular" signifies "tuberculoid."—EDITOR

were destroyed fairly early, in lepromatous cases they seemed to persist for a long time, although the normal numbers of fibers were rarely found. The fibers when found appeared healthy, though they often coursed through a focus of infiltrate. Hair follicles were seen which did not show any surrounding infiltrate or any nerve fibers ending in them. This would suggest that a secondary degeneration is responsible for the disappearance of these fibers.

That the hairs and their follicles play an important role in the reception of tactile stimuli has long been known. Shaving a normal part of the skin reduces to a large extent the sensitivity of the skin to touch (Larsell (11)). Our clinical observations on the changes in tactile sensibility in the different types of lesions undoubtedly show that the hairs play an important part in the retention of tactile sensibility by the leprous lesions. This sensibility has a direct relationship, not only to the presence or absence of hair on the lesion, but also on the amount of involvement of the hair follicle. While the presence of a few uninvolved hairs may still account for the persistence of tactile sensibility, the presence of even a few healthy fibers at the root may be enough to receive the tactile stimulus.

This latter proposition becomes even more apparent when it is realized that the hair follicle is supplied by nerve twigs coming from at least two different branches of the main trunk. Since it has been shown experimentally (Weddell (16)) that cutting off one of the supplying branches interferes with accurate localization and two-point discrimination, it would be interesting to study the different lesions of leprosy clinically with a view to ascertaining if any changes occur in accurate localization and discrimination. It would appear from the above findings that for experimental purposes it is of some importance to test all the lesions of leprosy for tactile sensibility before and after shaving. This, however, may not be of much practical value.

Anatomical studies have shown that the unit areas of sensory reception of the skin overlap each other, and that each sensory spot is innervated by endings derived from separate nerve fibers (Weddell). This is brought about by a complex intertwining plexus of nerve fibers below the epithelium, but there is no continuity between the overlapping fibers (Larsell). Considering this anatomical basis of tactile sensibility, it is conceivable that, even if a number of free nerve endings are destroyed by the disease, a few persisting fibers may still carry the stimuli. This may be particularly true of a very sensitive area like the face where, often, leprosy lesions fail to show change in tactile sensation.

While interpreting the changes in the subepithelial regions of the corium in the different types of lesions, one should keep in mind that in the macular and tuberculoid cases one is examining a piece of a localized lesion, while in the lepromatous case the piece is from a diffusely infiltrated skin. This brings us to the question of the evolution of lepromatous

leprosy. That the bacilli could have primarily invaded the fine nerve twigs in all the areas is difficult to understand, in view of the large numbers of apparently normal fibers found in the early stages of lepromatous leprosy.

The various changes in the axis cylinders described above bear a remarkable resemblance to those described as occurring in axons during Wallerian degeneration. From this it would appear that all changes in the fine cutaneous nerves in leprosy can be explained by the occurrence of Wallerian degeneration. Khanolkar (9) suggested that the bacilli travel along the axons and, according to Fite, the occurrence of bacilli inside the axis cylinder has been described by Lie, Uhlenhuth, and Takino. Fite himself has described the occurrence of bacillary masses distending the central canal of the nerve fiber. On the other hand, many have expressed themselves against this view. So far, we have failed in our efforts to demonstrate lepra bacilli in the sections stained for nerve fibers. Whatever may be the exact location of the bacilli in the nerve, it is unnecessary to assume that the changes in the fine cutaneous nerves are due to a direct action of the bacilli. The presence of apparently normal nerve fibers in the band of infiltrate in early lepromatous cases, in spite of their being surrounded by an ocean of bacilli, lends support to this contention. From our observations it would appear that in tuberculoid leprosy the marked inflammatory reaction which occurs inside the nerve bundles in the deeper corium and subcutis destroys the fibers rapidly by pressure, while in the lepromatous cases the process in the nerve is an apparently slower one, thus accounting for the persistence of the fibers for a long time.

The presence of a diffuse inflammatory infiltrate in a band-like form in lepromatous cases can easily be explained by a lymphatic spread of large numbers of bacilli along the extensive intercommunicating plexuses of lymphatics in the upper and middle parts of the corium, thus affecting large areas of the skin by continuity. Future investigations directed to the study of lymphatics of the skin in this disease may throw more light on its evolution.

SUMMARY

1. A study of the cutaneous nerves in leprosy lesions of different types is presented, and an attempt is made to correlate the clinical observations of tactile sensibility with the histological findings.

2. The importance of hair follicles in preserving tactile sensibility is stressed, and also the advisability of testing all types of lesions before and after shaving to determine this type of sensibility.

3. The damage to the nerves in the different kinds of lesions is of the same kind, but of different degrees. It is influenced by the nature and the age of the lesions.

4. The occurrence of fusiform swellings and bubbles along the course

of the axons, and of twisting, flattening, and fragmentation of axis cylinders, can all be accounted for by Wallerian degeneration, which probably is a consequence of damage by pressure on deeper nerve fibers.

RESÚMEN

Los autores presentan un estudio de los cambios histológicos en los nervios cutáneos en lesiones leprosas de varios tipos. Se observó gran correlación entre la presencia de folículos pilosos y sensación cutánea. Los cambios nerviosos fueron del mismo tipo aunque de variable intensidad en los varios casos estadiados, y son influenciados por la edad de las lesiones. Los recrecimentos fusiformes, fragmentaciones y distorciones se interpretan como debidos degeneracion Walleria, como consecuencia de daño causado por presión a fibras más profundas.

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DESCRIPTION OF PLATES

(All photomicrographs 460× except Figs. 3 and 8, which are 970×. Reproduced without reduction.)

PLATE (1)

FIG. 1. Showing separation of nerve fibers inside the nerve bundle, in a maculo-anesthetic lesion.

FIG. 2. Separation of nerve fibers and infiltration by inflammatory cells, in a tuberculoid lesion.

FIG. 3. Nerve fibers showing knotted appearance of the axons, in a tuberculoid lesion.

FIG. 4. Fragmentation of axons, in a tuberculoid lesion.

FIG. 5. Lepromatous lesion showing fine nerve fibers entering the epithelium.

FIG. 6. Apparently intact nerve fiber coursing through the inflammatory infiltrate of a lepromatous lesion.

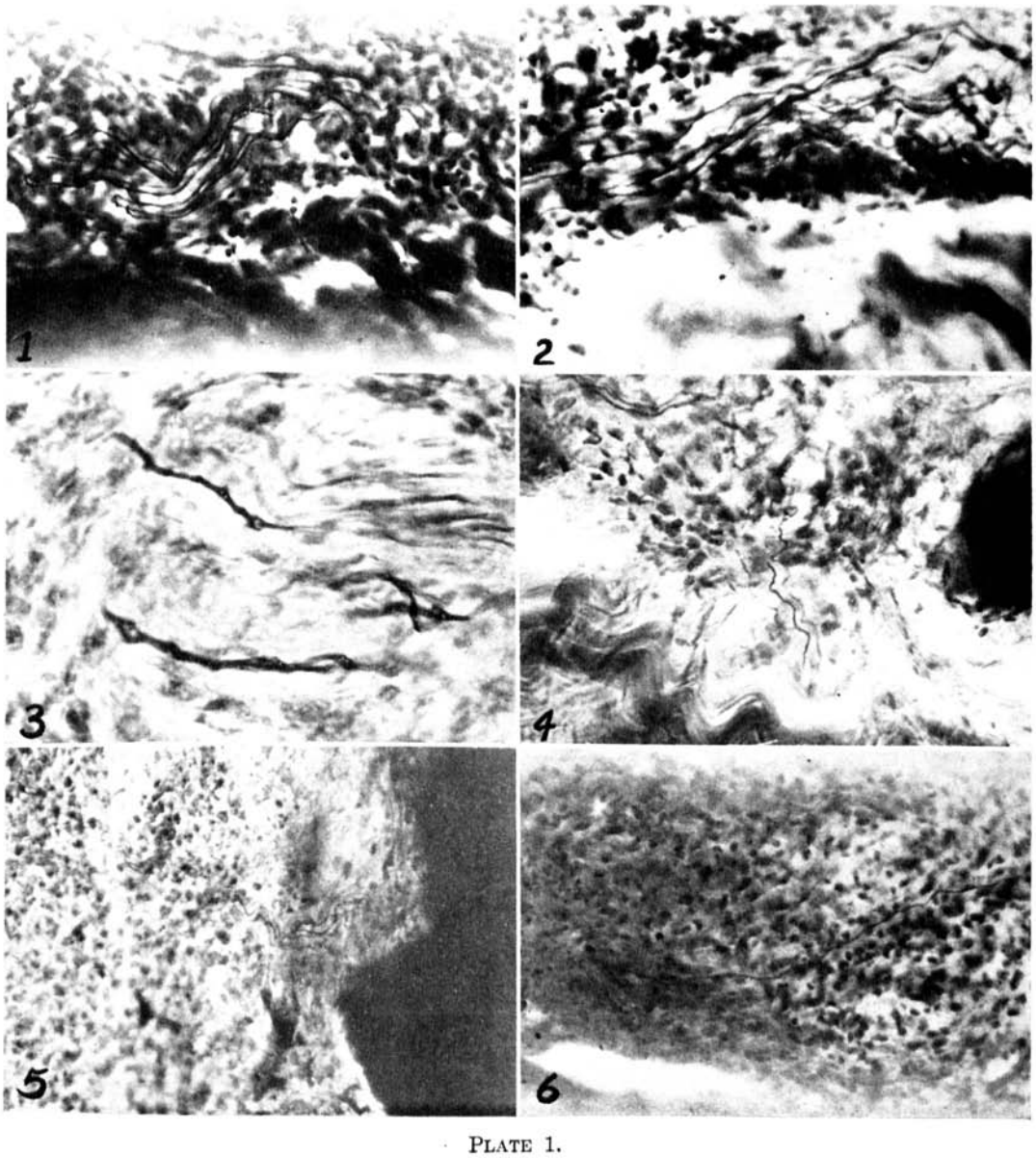

PLATE 1.

PLATE (2)

FIG. 7. Fragmentation of axons, in a lepromatous lesion.

FIG. 8. Fusiform swelling of the axons in a lepromatous lesion.

FIG. 9. Transverse section of a nerve bundle with thick perineurium, in a lepromatous lesion.

FIGS. 10 & 11. A nerve bundle in the deeper corium of a lepromatous lesion, showing paucity of axons and increased cellularity.

FIG. 12. A hair follicle showing two nerve fibers ending in it; lepromatous lesion.

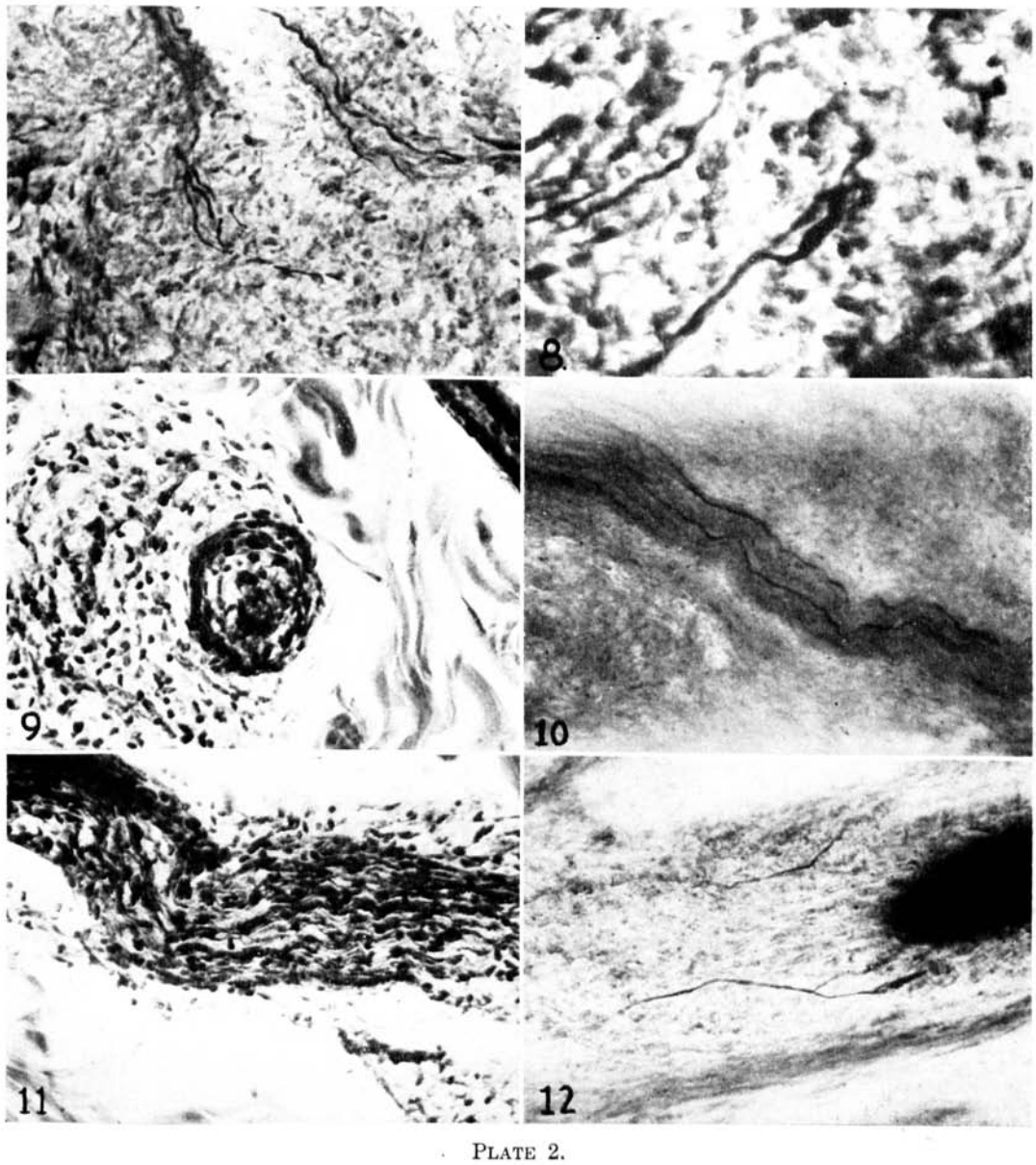

PLATE 2.