

# THE NERVE-CELLS OF THE HUMAN POSTERIOR ROOT GANGLIA AND THEIR CHANGES IN GENERAL PARALYSIS OF THE INSANE.

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ALTHOUGH there is a considerable amount of literature upon the changes in the cells of the posterior root ganglia in general paralysis of the insane, there still exists much diversity of opinion as to the extent to which those cells are affected, some authors describing advanced and destructive changes in the cells, while others are of the opinion that the affection is very slight.

It is worthy of note that before describing the changes to be found in general paralysis no one has attempted to give an account of the normal intimate structure of the posterior root ganglion cells; and it seems to us that a minute description of those cells, studied by the most modern methods now at our disposal, is an absolute necessity, because, as we hope to show, there are several types of cells which vary very markedly in size and in the arrangement of the chromophile elements, so that without a knowledge of the normal features one cannot attempt to describe the modifications which the cells undergo in the course of a disease such as general paralysis of the insane.

Early in our work it became evident to us that the older methods of fixation and hardening were utterly useless for the study of such a delicate cell as that contained in the spinal ganglia, and so we found it necessary to finally discard such fixatives as formalin and graduated alcohol.

As a result of adopting a new and reliable method of fixation, we have proved to our satisfaction that work conducted by the methods formerly used is full of artificially produced appearances, which in many cases have been mistaken for evidences of a morbid process. What those changes are we shall show before describing the changes found in the posterior root ganglia of general paralytics. But in the first place let us see how our knowledge stands with regard to the normal structure of the posterior root ganglion cells.

Before entering on a description of the types of cells found in the normal human posterior root ganglia we must refer to the work which has of late been done on the ganglia of the lower animals.

For several years it has been recognised that there were cells of different sizes in the posterior root ganglia, but, until comparatively recently, no attempt was made to classify the cells beyond the use of the terms "large" and "small." During the last five years, however, several observers have given some attention to the subject, and have classified the cells according to their size and to the arrangement of the chromophile elements within the cell.

Lugaro has summarised the literature on the subject and has given concisely the views held by various observers in his paper published last year; we shall refer to this fully later.

Dogiel, Ramon y Cajal and Oloriz recognised, when using Ehrlich's "*intra-vitam*" method, that there were large clear cells and small obscure cells.

Nissl, in 1897, described four types of cells in the posterior root ganglia of the rabbit; he distinguished them as follows:—

(1) Cells with large chromophile elements arranged concentrically around the nucleus.

(2) Cells having a single layer of elements arranged at the periphery, a thin clear zone just below this free from chromophile elements, and another clear space in the neighbourhood of the nucleus.

(3) Cells with a thin peripheral layer and a broader peri-

nuclear layer of large elements, and between these two a band of smaller elements, with sometimes some larger elements mixed with them.

(4) Cells somewhat similar to the preceding, but more or less indeterminate in character.

Van Gehuchten and Nelis, in 1898, distinguished the small and obscure cells from the large clear cells, and by the relative amounts of chromophile elements they distinguished numerous types which, they considered, were united by intermediate forms. They described the two types most commonly met with as being rich in chromophile elements, but in one these were large, irregular in shape, and scattered through the whole of the protoplasm of the cell; while, in the other variety, they were in the form of fine granulations uniformly distributed throughout the cell excepting a narrow clear peripheral zone.

Their third type included cells with a few fine irregularly shaped elements.

Next they described cells with large fusiform elements arranged concentrically around the nucleus.

They then mentioned a rare type in which a few large irregularly shaped chromophile elements were arranged exclusively around the nucleus; and, lastly, a still more rare type, whose elements were divided into two bands, one at the periphery and the other around the nucleus, and between the two there was a zone free from chromophile elements.

Cassirer in his description of the cells of the posterior root ganglia of the rabbit mentioned four types:—

(1) Large clear cells with the nucleus generally central, but sometimes displaced to the periphery; the elements in these were arranged around the nucleus, and at the periphery in concentric layers, and there was a narrow clear zone at the periphery, and a narrower one around the nucleus.

(2) Medium-sized cells, for the most part clear, with a large nucleus generally central, but sometimes excentric; around the nucleus there was first a zone of large chromophile elements, then one of very fine chromophile elements, and again at the periphery a zone of large elements.

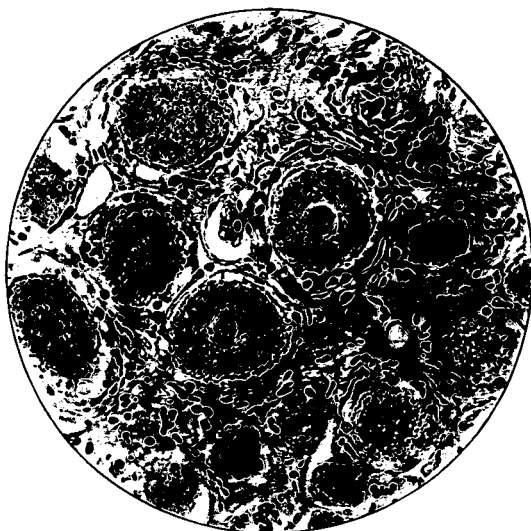


FIG. 1.

Two large clear cells, with central granuliform elements and a single peripheral ring of large chromophile elements. Note the perinuclear space. *Lugaro's type, No. 2.* Zeiss, apochrom., obj. 8 mm., oc. 4.

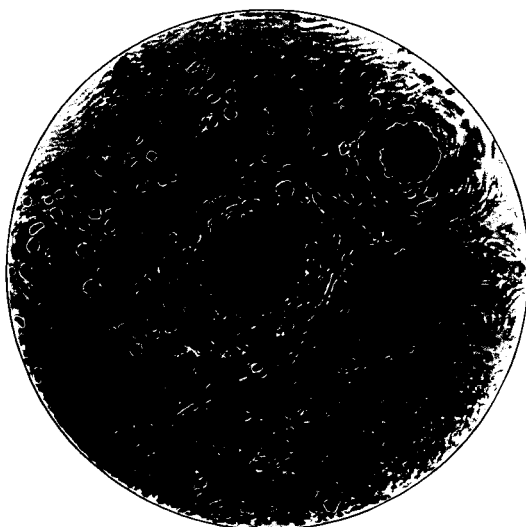


FIG. 2.

Early stages of chromatolysis in type 2. The nucleus is slightly excentric and the peripheral edge of the central granuliform mass is beginning to break up. Note the persistence of the peripheral chromophile ring. Zeiss, apochrom., obj. 8 mm., oc. 4.

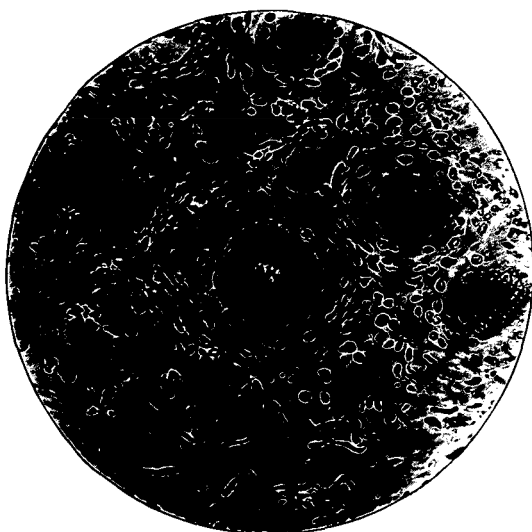


FIG. 3.

More advanced stage of chromatolysis in type 2. The central chromophile mass is very degenerated at its periphery and there is a clear space left between it and the large peripheric chromophile elements which are still well preserved. The nucleus here has retained its central position. Zeiss, apochrom., obj. 8 mm., oc. 4.

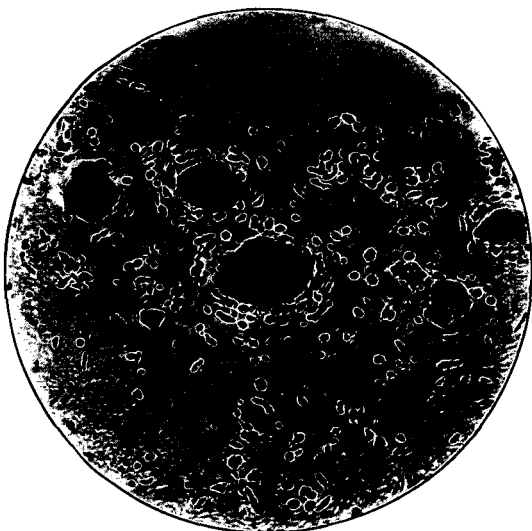


FIG. 4.

In this example of type 2 the central chromophile mass has disappeared with the exception of a few granules around the nucleus. Note that the peripheric chromophile ring still persists at this late stage of chromatolysis. There is a small dark-staining band of chromophile substance attached to the outer wall of the central side of the nucleus. Zeiss, apochrom., obj. 8 mm., oc. 4.

(3) Cells as large as the last, but of irregular shape, with a smaller nucleus, often excentric, and the elements indistinct because of the staining of the fundamental part of the cell.

(4) Small cells with a relatively large nucleus, with the elements arranged irregularly and with the fundamental part of the cell stained. Sometimes in these cells two nuclei were seen.

Cox also in 1898 published a paper in which he described the cells in the posterior root ganglia of the rabbit, but he differed from the above-mentioned authors in that he considered that the types of cells were clearly distinguished from each other, and he failed to find any intermediate forms between the various types.

He mentioned two types, and the first of these he divided into two sub-types, cells large and cells small.

In the large cells of type 1 the chromophile elements were not large, they were of irregular shape and only at the periphery did they assume an elongated form ; no concentric arrangement was seen excepting at the periphery and here there was only one layer so disposed. The nucleus was situated in the middle of the cell and was rarely double.

The small cells of type 1 were distinguished by the fact that the fundamental substance between the chromophile elements had a tendency to stain more or less deeply. The elements were distinct, but always much smaller than those in the larger cells.

The cells of type 2 had elongated chromophile elements arranged in layers, some layers were concentric around the nucleus, and others were concentric around the central point of the cell. The nucleus was always excentric, often markedly so.

He then referred to the different appearances which this type of cell presented in sections which were made in different planes, and showed that in one section the nucleus might be near the centre and in another placed at the extremity of the cell.

It will be seen that there are considerable differences

between the descriptions of these cells given by the above mentioned authors; these differences have been cleared away, and in fact the classification of the types of cells met with in the posterior root ganglia has been placed on a firm and scientific basis by Lugaro in his valuable paper published last year.

Working with a new and improved technique, which will be described fully later in this paper, he divided the cells in the posterior root ganglia of the dog into five types, quite distinct from each other and having no intermediate forms. His first type consisted of clear cells, the largest in the ganglion, with fine granuliform chromophile elements scattered diffusely throughout the cytoplasm, and only a little increased in size and density at the periphery, internal to a narrow limiting clear zone, which was entirely deprived of them; around the nucleus also there was a narrow clear zone which was free from any chromophile elements; the nucleus was large and clear and was situated in the centre of the cell (fig. 6).

In his second type he included clear cells of medium size generally, although some of them equalled those of the first type. In these cells the chromophile elements around the nucleus were a little larger and less crowded than in type 1; they were bounded externally by a zone of varying width in which there were few, if any, elements, and outside this there was a single layer of large elements. At the periphery and around the nucleus there was a well marked narrow clear zone. The nucleus was large and clear and was always central (fig. 1).

The third type Lugaro spoke of as "scure" or darkly-staining cells. Many of these were small, but others were quite of medium size; they were frequently irregular in shape. The chromophile elements were very minute in the central part, and slightly larger towards the periphery. The distinguishing feature of this type was that the fundamental substance of the cells was stained more or less deeply, and this was the more marked in those of larger size. It gave rise to the dark, undefinable appearance similar to that seen in partially differentiated cells.

Frequently there was considerable difficulty in defining the outline of the chromophile elements, owing to the deep staining of the fundamental substance. The nucleus also was deeply stained; it was central, and was not relatively so large as in the other types. The peripheral and perinuclear zones were present, but were less distinct than in the clear cells.

The fourth type consisted of small or medium sized cells, in which the chromophile elements were large, few in number, and regular in shape, and had numerous branches attached; they were arranged concentrically around the nucleus, and there was a well marked peripheral and perinuclear clear zone. The nucleus was large, clear, and was situated centrally in the cell (fig. 8).

His fifth type of cell, corresponding to the type 2 of Cox, included cells of medium and small size. Some of these were round, and others were decidedly oval in shape. The nucleus was constantly excentric, often markedly so. The chromophile elements were arranged in concentric layers around the central point of the cell; those at the periphery were large, cohering, elongated, and united to each other by fine branches; those in the central part were smaller and more irregular, but the concentric arrangement was maintained. The peripheral and perinuclear clear zones were quite distinct (fig. 10).

The above description is a picture of a section of one of these cells made in the direction of the long axis of the chromophile elements; when the section was made in a plane at right angles to this, the chromophile elements were seen to have an irregular shape, and they were connected by their branches so as to present a reticulated appearance; their size, the excentricity of the nucleus, and the concentric arrangement of the chromophile elements will assist one to recognise them.

Moreover, intermediate forms, in which the section was made obliquely to the long axis of the chromophile elements, were frequently seen. In some cells too, the nucleus was placed at one end of the cell, with layers of chromophile elements disposed concentrically around it, and at the other



pole of the cell there was a distinct spiral arrangement of the elements.

This cell clearly corresponds to the cell referred to by Lenhossek as being found in the frog, in which he spoke of this disposition of the nucleus and spiral as "centrosome" and "sphere," and he considered it a cell in process of reproduction.

Levi, however, in 1897, noticed a similar cell in the toad, and he showed that in this type of cell the nucleus, with concentric layers of chromophile elements around it, was normally situated at one pole of the cell, and that the other pole was occupied by a figure, which he described as a "spiral vortex of fibrils and chromophile elements."

The above are the five types of cells which Lugaro described in the posterior root ganglia of the dog.

In his article on the posterior root ganglia of the rabbit he included types 1 and 2 under one head, because he found that in the normal cells there were some points of similarity; and he also found that, in the course of their degeneration after section of the afferent nerve, they passed through the same definite series of changes, and only at the very commencement were they distinguishable.

At the beginning of our work on this subject, we examined the posterior root ganglia of lower animals with the view of confirming the findings of Lugaro, and we then directed our attention to the root ganglia in man.

We have cut sections of the posterior root ganglia of the cat, the dog, and the rabbit, and we can fully confirm the description of the types of cells given by Lugaro; and a careful examination of the root ganglia in man has shown that the same types of cells exist there also.

We have followed Lugaro's classification, because it is the most recent and the most complete, and because we consider that a comparison with all the descriptions given above might lead to some confusion.

Our normal specimens were obtained from cases of acute mania who died soon after admission into the asylum. We

feared that it would be impossible to eliminate the fallacies due to imperfect fixation and to *post-mortem* change, if we relied on tissues obtained from a general hospital.

### CELL TYPES IN THE NORMAL HUMAN POSTERIOR ROOT GANGLIA.

In the human posterior root ganglia we have been able to separate and to distinguish five independent varieties of cells, which show differences both in the normal arrangement of the chromophile elements, and in the way in which they undergo modifications in their degenerative phases.

The most common type of cell met with is that which corresponds to Lugaro's first type. The cells in this type are large and clear; the chromophile elements are granuliform, and are scattered throughout the entire cell body; but at the periphery there is a slight increase in their size (fig. 6). Outside the chromophile elements there is a well marked clear peripheral zone devoid of chromophile substance, and around the nucleus which is central, there is also a similar well defined clear zone, called the "perinuclear space."

The next type in frequency is that which corresponds to Lugaro's second type in the dog. We find this cell is clearly distinguishable from the preceding by the large size of the peripheral chromophile elements, which are arranged in a single ring around the cell (fig. 1). Internal to this ring the chromophile material is granuliform, the individual elements being very minute and irregular in size.

The granules are usually slightly increased in number around the centrally placed nucleus.

The peripheral and perinuclear clear zones are again prominent features, as in the first type.

The third and last type of cell, in which the chromophile material is exhibited in the form of granules, is that termed by Lugaro the "scurc" or darkly-staining cell. In the human subject we find a large and small variety of this type.

In the smaller cells the chromophile elements are very minute, with indistinct outline, and are scattered diffusely

throughout the cytoplasm. In the larger cells the chromophile elements are larger, and are arranged in two concentric condensations—one around the nucleus and the other towards the periphery. In both the fundamental amorphous substance stains deeply, and obscures to a large extent the chromophile material; the nucleus, which also stains deeply, is central. A perinuclear clear space is recognised with difficulty, but the peripheral clear zone can always be seen, though narrower than in the large clear cells.

The two remaining types, about to be described, are sharply defined from the preceding three varieties by the large size of the individual chromophile elements. The granuliform chromophile substance, which forms the main feature in types 1, 2, and 3, is replaced in types 4 and 5 by an element which is much larger and more distinct, but the shape and arrangement is not the same in the two types.

Under type 4 we find two varieties, one with small cells and the other with cells of medium size, and in these there are certain characteristics common to both. The chromophile elements are large and comparatively few in number; the fundamental achromatic substance is either unstained or only very slightly coloured; the nucleus is central, and the peripheral and perinuclear clear spaces, which we have mentioned as being common to all the types, are prominent features in these cells (fig. 8).

In the smaller variety the chromophile elements are smaller and a little more slender than in the medium-sized cells, and they are arranged concentrically around the nucleus.

In the medium-sized cells the chromophile elements are larger, and tend to assume a diamond or square shape, and they do not present such a complete concentric disposition as is seen in the smaller variety. Both varieties, therefore, of this type can be clearly differentiated from any of the preceding types by the large volume and the well-defined outline of the chromophile elements.

In the other type of cell, which corresponds to Lugarq's fifth type in the dog, the chromophile elements are as large as in type 4, but differ from them in their shape and in their

disposition around the nucleus. The nucleus is normally markedly excentric and frequently occupies a peripheral position, but it is always separated from the extreme edge of the cell by a narrow band of chromophile substance. The chromophile elements are long and slender in the large examples of this type (fig. 10), and they radiate from the nucleus in successive rings towards the opposite pole of the cell. The elements are more closely packed together around the nucleus, but further away the space between adjacent rings is greater. In the smaller vorticose cells the elements are a little thicker, relatively to their length, than those in the larger variety, but the same arrangement in the cell prevails.

Sections of these cells made in different planes exhibit the same pictures as those described by Lugaro in the cells of the dog.

Various authors who have written on the subject of the posterior root ganglia have drawn attention to the fact that the size of the cells varies considerably in the ganglia of the different regions of the cord, the great majority of cells in some ganglia being large and in other ganglia small; and they have suggested that as the cells differ in size in various ganglia, their function also must be distinct. We have found that the cells do vary in size in different ganglia, but in each ganglion every type of cell is present, and there can be no doubt that in the ganglia in which the cells generally are small, those cells which belong to the smaller types are reduced in size, as well as those which belong to the larger types, and we must assume that the same type of cell has the same function in every case. It is hardly correct, therefore, to say that the large cells are absent or relatively greatly reduced in number in certain ganglia.

Another source of fallacy lies in the fact that in certain ganglia almost all the cells along one side may be of large size. If a section was made along this side, practically only large cells would be seen; but if a section was made at right angles to this, the various types would be seen in their usual proportions. We have, therefore, found it necessary to cut sections right through the ganglion.

## THE PERINUCLEAR SPACE.

In the above description of the cell types, reference has been made to a perinuclear space, which forms one of the constant histological appearances met with in the nerve-cells of the posterior root ganglia.

This perinuclear space is worthy of special mention, in view of the importance now attached to the intracellular lymphatic system and its connections, as there seems to be a large amount of evidence to show that this space forms part of the canalicular system described by Donaggio, Holmgren, and others.

It is not our intention to enter into a description of the lymphatic canalicular system or of the endocellular vessels (Fritsch, Holmgren, and Adamkiewicz), as such is quite outside the scope of this paper; but we may be pardoned for dwelling on the perinuclear space a little more fully, as it might be questioned whether the appearance was really a normal feature of the cell, or an artefact due to the effects of hardening or shrinkage.

Below it will be seen that considerable trouble has been taken to select the best fixative for these delicate structures in order to eliminate as far as possible faulty fixation or shrinkage during the subsequent operations of hardening and embedding. But first let us look at the opinions to-day advanced as to the precise nature of this structure.

In connection with their work on the endocellular lymphatic canalicular system, Holmgren and Donaggio have observed and described a distinct perinuclear space, containing no chromophile material, into which the canaliculi appear to open, and Holmgren seems to consider that the perinuclear space is to be regarded as the centre of the endocellular circulatory paths. We have ourselves observed two or three of these canaliculi in some cells of the first type, but we have not yet seen any instances where they open into the perinuclear clear space. The canaliculi are seen as clear, sinuous bands, surrounded on both sides by chromophile elements. Confirmatory evidence of the existence of this structure is to be

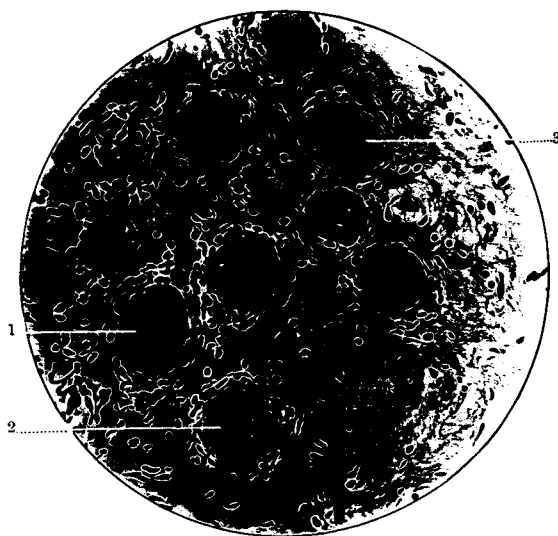


FIG. 5.

Note three cells of type 2 showing—(1) Early chromatolysis with excentricity of nucleus; (2) Degeneration of periphery of central granuliform mass; (3) Disappearance of central chromophile elements and persistence of peripheric chromophile zone. Zeiss, apochrom., obj. 8 mm., oc. 4.



FIG. 6.

A large clear cell with granuliform chromophile elements. There is a slight increase in size of the elements towards the periphery but no definite prominent peripheric ring as in type 2. *This is Lugaro's 1st type.* Note the perinuclear and peripheral clear spaces. Zeiss, apochrom., obj. 8 mm., oc. 4.



FIG. 7.

Two large clear cells of type 1. In one there is a small localised deposit of pigment at one extremity of the cell; in the other there is a diffuse chromatolysis with marked excentricity of the nucleus. Zeiss, apochrom., obj. 8 mm., oc. 4.



FIG. 8.

Medium-sized cell with large chromophile elements. Note the difference in size between the elements in this type and types 1 and 2. This is *Lugaro's 4th type*. Leitz, obj. 7, oc. 1.

found in the embryological researches of Fragnito, Colucci, Capobianco, and Piccinino, who, in working at the development of the nerve-cell, lay special emphasis upon the peculiar mode of development of the perinuclear space and the canaliculi.

Without going completely into a consideration of these authors' work, it is sufficient for our present purpose to note that they demonstrate the pluricellular origin of the nerve-cell, and in the course of their work they show how in the process of development the perinuclear space is formed, and that it is present at an early period of the existence of the cell.

The above-mentioned authors are agreed that the nerve-cell is formed from a primary neuroblast and several secondary neuroblasts, and that each neuroblast is surrounded by protoplasm.

The primary neuroblast becomes the permanent nucleus of the adult cell, while the latter become associated with it and undergo certain modifications to form the adult cell-body with its contents. Fragnito then shows that between the primary neuroblast and the secondary neuroblasts there is a space, which becomes the perinuclear space, and that this communicates with the spaces between the secondary neuroblasts, which become the canaliculi of the adult cell. In support of Fragnito's observations, Colucci affirms that the protoplasmic substance surrounding the primary neuroblast presents no true continuity with the nuclear wall. There is here a clear interposing ring in which there is a very faint, refractile, substance. As the surrounding protoplasm grows, the more evident does this clear ring become.

There can be little doubt, then, that the perinuclear ring is not an artificially produced appearance. It may be suggested that the reagents used in fixation and hardening caused retraction of the chromophile substance from the nuclear wall: but the observations of Colucci were made on fresh as well as hardened nervous tissues, stained with aniline dyes, and he found the same appearance in both instances.



## PERIPHERAL CLEAR RING.

There remains yet to be noted the peripheral clear ring, which also forms one of the histological features of all the cells types mentioned above. This ring varies in width according to the type of cell, being larger in the large cells and smaller in the small cells.

The chromophile elements do not reach the margin of the cell body, but terminate abruptly, leaving a clear zone around the entire cell. This is especially well seen in types 1 and 2. The outer margin of this zone is regular in outline and stains slightly more deeply, and so gives to the cell a well-defined edge and separates it sharply from the surrounding capsule.

In this peripheral clear zone no trace of chromophile substance can be seen, but minute fibrils are observed, with high magnification, to traverse it in all directions, giving one the impression of a fibrillar network. Between the external margin of the peripheral clear protoplasmic ring and the capsule enclosing the nerve-cell, there is a narrow pericellular space, crossing which one observes numerous minute, faintly-staining threads, which apparently run from the surrounding tissues into the peripheral clear zone of the cell body.

As to what these thread-like structures are, we hesitate to give a decided opinion; but the observations of Holmgren, Studnicka, Donaggio, and others, lead us to suggest that they may be of the nature of vascular or lymphatic structures.

Seeing that we have not employed any special technique or staining to warrant us in giving any definite view on this subject, we at present merely make a statement of their existence, while describing the peripheral clear ring which surrounds the ganglion cell.

## FIXATION, HARDENING AND EMBEDDING.

The choice of a suitable fixative and the subsequent hardening in alcohol are of the utmost importance in dealing

with such delicate structures as the ganglion cells under consideration.

It is interesting to note first the researches of Lugaro in this connection, and his results.

The following solutions were employed by him :—

(1) Equal parts of a saturated solution of corrosive sublimate and picric acid.

(2) Mann's solution (sublimate solution, picric acid and formaldehyde).

(3) Carnoy's fluid.

(4) Gilson's fluid.

(5) Nitric acid (5 per cent.).

(6) Cox's first mixture (saturated sublimate solution, 30 parts; osmic acid, 1 per cent., 10 parts; acetic acid, 5 parts).

(7) Cox's third mixture (saturated sublimate solution, 30 parts; formol, 10 parts; acetic acid, 5 parts).

The results were as follows:—In Cox's fluids some shrinkage occurred, while Carnoy's fluid fixes the centre better than the periphery of the cell. The mixture of saturated sublimate solution and picric acid, Mann's fluid and nitric acid gave the best results, not a single cell showing the least appearance of shrinkage.

In staining, Lugaro found toluidin blue the best stain, and in the process of differentiation he recommended absolute alcohol in preference to any essential oil. An accessory stain, such as erythrosin, he found to be harmful in its action, because it took place at the expense of the chromophile elements, the smallest of which tended to disappear. The selective properties of the stain differed with the fixing agent; with picric acid and sublimate and with Mann's fluid the smallest chromophile elements were projected with incomparable clearness on a transparent background, with nitric acid the colour was paler and the contour of the chromophile elements less distinct. Carnoy's and Gilson's fluids gave good results, but they were inferior to the others named. Delafield's hæmatoxylin gave good results after fixation in picric acid and saturated sublimate solution, and in Mann's fluid. We find that it is necessary to use this

stain in dilute solution (1-80), and good results are obtained after one hour's immersion.

In our own work we have used as fixatives,

- (1) Mann's fluid.
- (2) Saturated sublimate solution (Heidenhain).
- (3) Saturated sublimate solution and picric acid.
- (4) Nitric acid, 5 per cent.

We have found that the mixture of saturated sublimate solution and picric acid in equal parts gives us the best results with toluidin blue staining, although it may be mentioned that saturated sublimate solution alone is also a very good fixative.

It is impossible to attach too much importance to the special fixation and embedding methods which must be employed for the cells of the posterior root ganglion, because without such special fixation and staining, a recognition of the varieties or types of cells which comprise the ganglion is quite impossible.

The mode of preparation of the human posterior root ganglion, which we have finally adopted, after a somewhat lengthy trial, is as follows:—The ganglia to be studied must be obtained from the cadaver before *post-mortem* changes have had time to affect the cells even in the slightest degree. It can easily be understood that no matter how good the fixative employed may be, if *post-mortem* changes have set in an accurate picture of the nerve-cells cannot be obtained, and it can readily be demonstrated that the earlier the tissue is fixed the more do the results approximate to the appearances observed in animals whose ganglia have been fixed immediately after death.

The ganglion, which is carefully separated from the cord, is split into two pieces with a sharp razor, as we find that, without such an incision, penetration by the fixative is greatly delayed on account of the density of the capsule surrounding the ganglion and nerves. The division of these structures materially facilitates the rapid fixation which is so necessary, and allows the sublimate and picric acid to penetrate the ganglion from both periphery and centre.

The tissue is allowed to lie in the solution for three days—a less time scarcely suffices for the fixation of the larger ganglia; at the end of this time the fixative is poured off, and the process of hardening in graduated alcohol, commencing with 40 per cent., begins. When the hardening is completed the tissue is at once embedded in celloidin in the usual way, because we consider it preferable to keep the tissues in celloidin rather than to allow them to undergo prolonged immersion in alcohol. When required for sectioning, the embedding is completed by immersion in origanum oil and paraffin (equal parts) for twelve hours at a temperature of 45° to 50° C.; finally in paraffin alone for twelve hours, care being taken to reduce the deleterious effects of over-heating in the bath to a minimum.

The combined method of celloidin and paraffin embedding has been employed because it serves the double purpose of retaining the cells in position and at the same time allows of the thinnest possible sections.

It is necessary, in our opinion, to use celloidin as otherwise the cells may fall out of the section, leaving empty spaces, an appearance which will be referred to later.

We have laid special stress on the above technique because it seems to us that faulty fixation is entirely responsible for the spaces and shrunken cells, often described as signs of morbidity. Such so-called morbid appearances are very easily obtained in the posterior root ganglia of healthy lower animals, if one allows *post-mortem* change to become established, or if an inadequate fixative, such as alcohol, is alone used.

Tissues from the healthy animal have been treated by the various solutions mentioned above, as well as morbid tissues from the insane, and whereas the healthy cells, where faulty fixatives were used, exhibited a considerable degree of shrinkage, the cells in the morbid ganglia, treated by the most recent methods of technique, showed no sign of such an appearance.

When shrinkage of the nervous tissues does occur, whether from imperfect fixation, *post-mortem* change, or both combined, the appearances obtained are constant and

typical, and, as will be shown later, do not resemble in the slightest degree the pictures of true chromatolysis.

The first portion of the cell to be affected by the shrinking process is the peripheral clear zone previously mentioned. The pale achromatic substance forming this structure first loses its definite outline and deep indentations appear on its edge; as the change progresses, these indentations increase, until they reach the external limits of the chromophile part of the cell. On this stage being reached the cell is found to be without a proper peripheral clear ring, and this structure persists only in thick and thin deep staining bands uniting cell with capsule, and bridging across the artificially enlarged pericellular space.

In our description of the normal clear peripheral zone, it will be remembered that we described fine pale threads passing across the pericellular lymph space, and forming the only evidence of anatomical continuity between the nerve-cell and its capsule. It seems to us that it is along those threads that the cytoplasm shrinks, collecting on them, and thus giving rise to the thickened bands attached to the tissue surrounding the cell; it is only in this manner that we can see our way to account for the disappearance of the pale clear peripheral zone, which we know exists in the normal unshrunk cell, and for the development of the bands which occupy the former position of the peripheral protoplasmic ring. In consequence of this shrinkage the staining processes also are very much interfered with, the elements take the dye in a different manner, and the recognition of the cell types becomes a matter of no small difficulty.

With a more severe degree of shrinkage the bands of altered cytoplasm are prone to rupture, and on manipulation of the section, the cell drops out of its capsule leaving a clear open space. Such an appearance we have not yet seen as a result of a true degenerative process, and we hope to show that as the cell undergoes disintegration, its site is gradually filled up by proliferation of the surrounding connective tissue nuclei.

It seems to us therefore that unless *post-mortem* change

and artificial shrinkage are entirely eliminated, the existing true pathological processes are to a very large extent masked, and artefacts or pseudo-pathological processes receive an attention sufficiently great to lead to a grave source of fallacy in the estimation of the extent of the morbid change present.

#### MORBID CHANGES IN THE CELLS OF THE POSTERIOR ROOT GANGLIA IN GENERAL PARALYTICS.

It has been necessary to enter into a lengthy description of the cell types in the normal posterior root ganglia, for the reason, that each type degenerates in a specific manner distinct from the others, and a knowledge of their normal structure is therefore necessary, in order to recognise each type at an early stage of degeneration, and to follow it through its progressive phases. This we have done in six typical cases of general paralysis, uncomplicated by tabes, only examining such ganglia as we were able to secure before *post-mortem* change had set in. In every case ganglia were taken from the cervical, dorsal and lumbar regions, and having been prepared according to the method recommended above, were stained by Lugaro's modification of Nissl's method. Toluidin blue being used in preference to thionin.

The chromatolysis revealed by this method must naturally be described under each of the five types of cells existing in the ganglia.

Of the two varieties of large, clear cells which are described as No. 1 and No. 2, the former persists practically unaltered, much more frequently than the latter. Chromatolysis is shown in the first type by a general breaking up of the granuliform elements throughout the cell, and by an early peripheral displacement of the nucleus. The elements around the nucleus often show a tendency to preserve their shape for a considerable time after those in the other parts of the cell have disintegrated. A cell in an advanced stage of degeneration shows its altered chromophile elements in the form of a fine dust scattered throughout the cell body (fig. 7).

The second type of cell, though structurally somewhat closely resembling the first type, is easily identified owing to the characteristic peripheral chromophile band which persists to a very late period of degeneration. The seat of the primary stage of degeneration is much more local than in the preceding type.

It will be remembered that the centre of the cell body is occupied by a granuliform mass of elements, and it is at the periphery of this mass, that is, immediately internal to the peripheral chromophile ring, that we have observed the process to begin (fig. 2).

The disintegration proceeds progressively towards the nucleus, and at the same time the peripheral large chromophile elements are seen to become united by their extremities to form a complete ring around the cell (fig. 3). With the progression of the degeneration, the central granuliform mass gradually disappears, until in the later stages the cell is seen to possess a well-stained nucleus and a peripheral chromophile ring only, the two being separated by clear, unstained cytoplasm (fig. 4). Occasionally, when the chromatolysis has reached this stage, a small, diffuse, chromophile mass is seen attached to the nuclear wall (fig. 4), and when the nucleus is excentric, the mass is present only on the side of the nucleus which is turned towards the centre of the cell.

During the progress of the chromatolysis the nucleus may pass to the periphery of the cell, but very frequently it remains central or nearly so.

In the "scurc" cells (type 3), with the onset of chromatolysis, the nucleus passes towards the periphery and the centre of the cell becomes clearer owing to the breaking up of the chromophile elements. The degeneration spreads until chromophile elements are recognisable only at the periphery, and the remainder of the cell stains diffusely blue.

The cells at this stage somewhat resemble the cells of type 2 in an advanced stage of degeneration, but they are much smaller in size, and do not stand out so clearly from surrounding structures, and the nucleus is much more

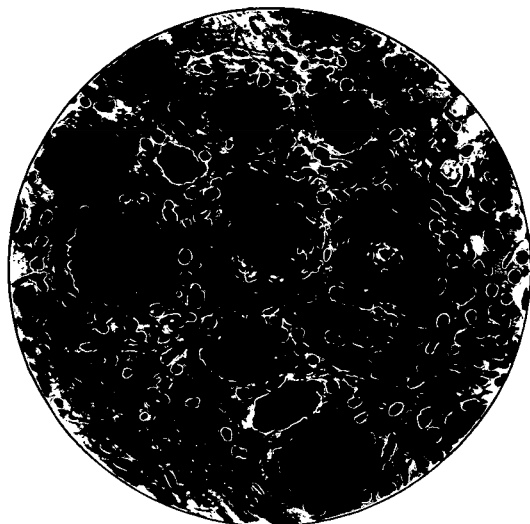


FIG. 9.

Note the cell in centre of the field. The nucleus is excentric and in its vicinity the chromophile elements are large. At the opposite end of the cell the chromophile material is broken up into fine granules. This is a degenerating No. 4. Zeiss, apochrom., obj. 8 mm., oc. 4.

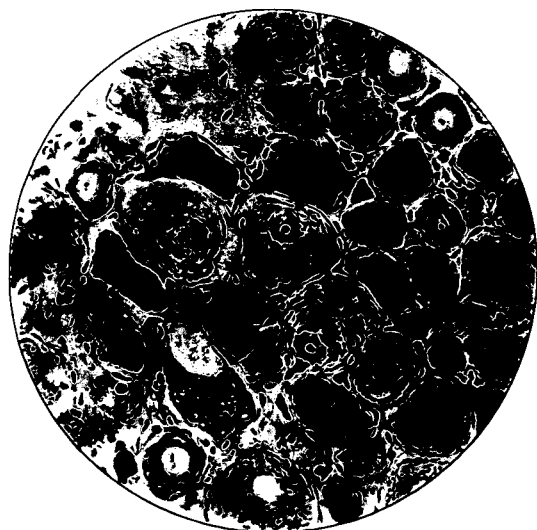


FIG. 10.

Two cells in centre of the field with elongated chromophile elements and excentric nucleus. Those are Lugaro's 5th type, and called by him "the vortucose cell." Zeiss, apochrom., obj. 8 mm., oc. 4.





FIG. 11.

Note cell with excentric nucleus, attached to which there is a chromophile condensation which points towards the centre of the cell. At the opposite pole of the cell the chromophile elements are breaking up. This is a degenerating vorticose cell. Zeiss, apochrom., obj. 8 mm., oc. 4.

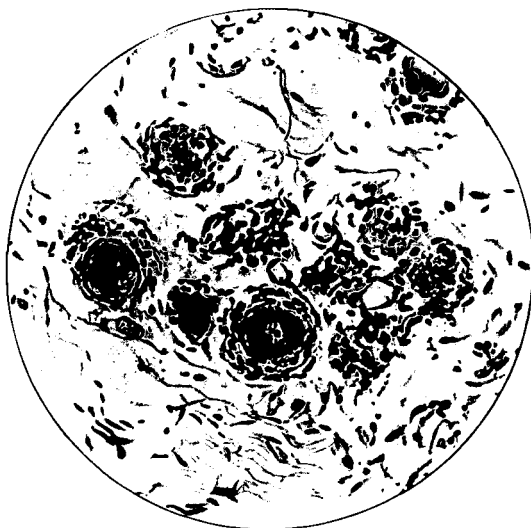


FIG. 12.

There are two degenerating cells around which can be seen a proliferation of nuclei. Above those two cells aggregations of nuclei are seen occupying the former site of degenerated nerve-cells. Zeiss, apochrom., obj. 8 mm., oc. 4.

frequently displaced to the periphery than in the large clear cells.

In type No. 4 the chromatolysis may commence at one focus in the cell, or may have a diffuse character from the beginning. The large elements are seen to become broken up into small, irregular granules, and the nucleus tends, at an early stage, to assume an excentric position, and when it has reached a point quite close to the periphery the largest remaining elements are observed in its immediate vicinity (fig. 9), while those in the other parts of the cell are represented by a fine diffusely staining dust.

This type of cell is always recognisable in its later phases of degeneration by the prominent, thick, chromophile elements placed between the sides of the excentric nucleus and the edge of the cell (fig. 9).

It has been difficult in the last type, that is the vorticoso cell, to follow out the morbid changes, because of its comparative rarity. We have examined numerous sections from the ganglia of general paralytics, and from cases of acute insanity, and we have been struck by the scarcity of this type of cell. Dr. Eades, who is at present working at the changes in the posterior root ganglia of epileptics, confirms this observation, having himself noticed how infrequently this type of cell is met with. We have, however, discovered cells in various phases of degeneration, and in them we have noticed that the chromatolysis commences at the periphery by a breaking up of the elements, and at the same time there is frequently a large condensation of deep staining chromophile substance attached to the side of the nucleus which is turned towards the centre of the cell (fig. 11).

The nucleus maintains its peripheral position, and at its sides some slender, elongated elements remain practically uninjured. These elements are no longer seen in the other parts of the cell. The elements which do remain intact are never so thick or so prominent as those seen in the cells of type 4 at a similar stage of degeneration, so that an examination of the elements around the excentric nucleus will still reveal the type to which the cell belongs.

Although it has been possible to classify the stages of

chromatolysis of the various ganglion cells under the above-mentioned types, we have seen a few cells which have become so disintegrated as to be beyond such classification.

When cells reach a more advanced stage of degeneration than that described above, we have noticed that the surrounding connective tissue nuclei proliferate and form a dense ring around the cell residue, encroach upon it, and gradually fill up the site previously occupied by it (fig. 12). Such extensive forms of degeneration, with nuclear proliferation around, occur with great infrequency, and it is only around cells showing this advanced stage of disintegration that any degree of nuclear proliferation is found, and moreover, it progresses *pari passu* with the destruction of the cell, until finally there is only a group of proliferated nuclei left to indicate the position which the ganglion cell originally occupied (fig. 12).

Notwithstanding the fact that so many ganglion cells show a considerable degree of morbid change, yet it would be quite erroneous to assume that those cells were dead, seeing that their nuclei not only maintain their regular outline, but also show a normal affinity for the staining reagent. Such being the case, we regard them as still capable of regeneration, a fact which is borne out by the experimental work of Lugaro and others.

His work on the cells of the posterior root ganglia in the dog and rabbit after resection of the afferent nerve proves conclusively that, although the chromophile elements may almost entirely disappear, yet complete restoration is possible, and the normal appearance of the cell may be re-established on the completion of the process of repair.

In our sections we have frequently noticed appearances which suggest an attempt at regeneration of the chromophile material taking place in the cell.

When the breaking up of the chromophile elements has reached its height, we have found not infrequently a small chromophile mass attached to the outer side of the nuclear wall which looks towards the centre of the cell, and in examples which may be taken to show more advanced stages of regeneration, this mass may reach a considerable

volume, occupying a large part of the centre of the cell, and Lugaro has shown that it is from this chromophile mass that the new Nissl-bodies are formed by diffusion and reconstruction of the chromophile granules.

Such appearances, coupled with the fact that the nucleus of the nerve cells invariably remains well stained, and persists in an apparently healthy condition until the latest stages of chromatolysis seems to explain to us why there are so many cells found showing the various stages of degeneration which we have described, and why there are so few cells which go on to complete destruction.

Granted that the nucleus does not die, then regeneration is possible on the removal of the cause of the morbid change; and it can hardly be doubted that the cause in this instance is the presence of toxins, which are now admitted by all to be present in the blood of general paralytics.

But it is conceivable that the toxicity which affects the tissues in this disease, is not constant in its intensity, but is liable to waves of remission or intermission during which the affected tissues, if not too severely injured, can recover, and this fact is borne out by clinical evidence.

It can easily be understood, then, how it is that we have found so few examples of the destruction of cells in the posterior root ganglia of general paralytics, and we believe that the cells of this system become affected by the action of toxins upon them, but that on the removal of those toxins, the cells are capable of regeneration, and that probably they may react to the poisonous effects of those toxins and become repaired more than once in the course of the disease, before their resistive power is finally overcome, and they undergo complete destruction.

We have shown that many cells become chromatolysed in consequence of the action of toxins upon them, but the chromatolysis stops short of actual destruction of the cell, leaving the nucleus apparently unimplicated. Recovery being still possible, it can readily be understood that the central prolongation of those cells ought not to undergo a secondary degeneration; and as a matter of fact repeated examinations show that they do not. The posterior nerve

roots examined by the Marchi and Weigert-Pal methods, even in teased preparations, present a very slight degree of degenerative change, and this is not to be wondered at, when one considers how few ganglion cells show extreme phases of chromatolysis such as would lead one to suppose that they were dead.

In this connection one cannot help noticing how frequently one finds an amount of fibre degeneration in the posterior columns of the cord, which cannot be accounted for by the degree of degenerative change in the posterior root ganglia. We acknowledge that here we are at variance with Sibelius and Vyrubow, who affirm that the degeneration in the posterior columns depends on the destruction of the posterior root ganglion cells. A glance at their description of the alleged morbid appliances reveals the fallacies contained therein, as they base their conclusions on such descriptions as a granuliform degeneration of the chromophile elements—although as we have pointed out a granuliform condition of the chromophile elements is a normal feature of types 1, 2, and 3—and a paucity of cells revealed by the fact that there are clear spaces in which no cell structure can be detected. Such pictures as the latter can never be accepted as indices of morbidity, as we have demonstrated that *post-mortem* change and faulty fixation can account for them entirely.

In addition to this, experience has shown us that as the cell goes on to complete destruction, its site becomes gradually occupied by proliferation of connective tissue nuclei. Even in acute insanities, where one finds a much more rapid form of chromatolysis in certain cells, although the chromophile substance may be wanting almost throughout the entire cell, yet the cell retains its normal shape and preserves its proper relationship with the surrounding structures.

As we have indicated before, spaces and shrinkage are artefacts, and if discounted from Sibelius and Vyrubow's work, the amount of fibre degeneration will assume a preponderance greatly in excess of the true degenerative change in the ganglia, and the results of those authors' research will more nearly approximate to those found by us.

We are not alone in considering that the changes in the posterior root ganglia are insufficient to account for the changes in the posterior columns, as Campbell and Fürstner have found little abnormality in the ganglion cells beyond a tendency to hyperpigmentation. Rubaud supports the above view, and considers that the scattered degeneration of the posterior columns occurs secondarily to an affection of the cells in Clarke's column. It seems to us more likely, however, that the diffuse degeneration found in the posterior columns in general paralysis uncomplicated by true tabes, is due to the toxins in the circulation which are capable of attacking the fibres everywhere in their course throughout the cord.

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