Acta neuropath. (Berl.) 16, 271—284 (1970)

Originalarbeiten · Original Investigations · Travaux originaux

The Ultrastructure of Medulloblastomas

F. Matakas, J. Cervós-Navarro

Institut für Neuropathologie im Klinikum Steglitz der Freien Universität Berlin

F. Gullotta

Institut für Neuropathologie der Universität Bonn

Received March 26, 1970

Summary. 9 medulloblastomas were investigated by electron microscopy. They all showed a rather unique histological and cytological structure. The cells were differently shaped, had in general many cytoplasmic processes and only few organells. Special differentiations which would have made tumour elements look like glial or neural cells were not observed when the tissue samples under investigation were carefully selected by light microscopical examination. Glial or neural elements were found only in infiltration zones where no clear distinction between pre-existing cerebellar and tumourous tissues could be made by conventional histological investigation. Medulloblastomas have essentially the same appearance as parvicellular sarcomas or embryonic tissues. Their common characteristic, viz. that they usually show no special differentiation, is also the most specific quality of medulloblastomas.

Zusammenfassung. 9 Medulloblastome wurden elektronenmikroskopisch untersucht. Sie zeigten eine relativ einheitliche histologische und cytologische Struktur. Die Tumorzellen haben eine wechselnde Form, wenig Organellen und in der Regel zahlreiche cytoplasmatische Fortsätze. In Gewebspartikeln, die nach der vorherigen lichtmikroskopischen Untersuchung aus dem Tumorzentrum stammten, wurden keine Zellen mit glialer oder neuronaler Differenzierung beobachtet. Nur in den am Tumorrand gelegenen Infiltrationszonen, wo die Tumorgrenze nicht mehr sicher zu bestimmen war, fanden sich gliale oder neuronale Zellelemente. — Das Medulloblastom hat die wesentlichen morphologischen Kriterien der kleinzelligen Sarkome und des embryonalen Gewebes. Deren gemeinsames Charakteristicum, daß sie nämlich keine speziellen Differenzierungen besitzen, ist auch eine spezifische Eigenschaft des Medulloblastoms.

Key-Words: Medulloblastoma — Electron Microscopy — Arachnoid Sarcoma — Lymphocyte-Like Cells — Dark Cells — Histogenesis.

Due to the fact that medulloblastomas infiltrate the cerebellar tissue it is difficult to determine which of the tissue structures are tumourous or autochthonous. This difficulty becomes even more complicated, when the tissue areas under investigation are minute as to the degree necessary for electron microscopy. As the tumour usually has a soft consistency, the surgeon removes most of it by suction. Thus in most cases the parts which remain for morphological investigation are mixed with infiltrated cerebellar tissue.

The only way to surmount these difficulties is by a selection of the tissue parts that are to be investigated electron microscopically by preparing broad sections of the epon embedded material for preceding light microscopical examination. Except for Voigt (1968) and in some respect for Waga (1965) the reports pertaining to the fine structure of medulloblastomas (Luse, 1962; Raimondi, Mullan, and Evans, 1962; Raimondi, 1966; Escourolle and Poirier, 1967; Sawada, 1967; Zülch

and Wechsler 1968) published so far, do not explain the criteria by which the tissue samples, that were supposed to reveal the structure of medulloblastomas, were selected. Consequently our observations differ in some respect from earlier investigations.

Material and Methods

For light microscopy, parts of the tumours in the vicinity of the fragments used for ultrastructural study were embedded in paraffine and stained with routine methods and by silver impregnation techniques. For tissue culture tumour fragments were explanted in roller tubes according to the method of Kersting (1961).

We investigated 9 medulloblastomas. Immediately after removal pieces of 7 tumours were fixed in 5% glutaraldehyde for 3 hours, washed in buffered saccharose solution for 1 to 4 days, fixed in osmium tetroxide for 2 hours, dehydrated in acetone, and embedded partly in Vestopal-W, partly in epon. In 2 tumours, which were treated as above after a formaline fixation of 4 years, the fine structure was not well preserved. However, the tumour specific qualities observed in properly fixed tumours could be recognized in these cases, too.

In order to get information about the general histological structures built up by the tumours, and to distinguish between tumour cells and infiltrated tissues semi-thin sections were made and stained with different methods (Matakas, 1969) before cutting the blocks on the ultrotome.

Results

One tumour had infiltrated the arachnoid and subarachnoid space. The number of mitosis and pyknosis phenomena was as great as it is usual in medulloblastomas. Only one tumour showed necrosis to any considerable extent. Collagen was demonstrable in all tumours. It was dispersed in fine fibres, except for a few smaller regions where it had accumulated in rather large bundles. The reticulin fibres were dispersed in "strands" and "fields" (Gullotta, 1967). There was no myxomatous degeneration, but in 6 tumours fat vacuoles were observed in a few cells. Histochemical investigations proved it to be neutral fat. In 3 tumours small areas with hyaline degeneration were present.

In the tissue culture the cell colonies consisted of small fusiform or triangular cells with oval nuclei that always contained a prominent nucleolus. The cytoplasmic body was small. The cells grew uniformly without any special arrangements. The cultures were full of small cells with hyperchromatic nuclei. While some of the cultures reached a steady state after a short period of proliferation, slowly proceeding to degeneration, others went on growing and proliferating without displaying an endothelial appearance so characteristic of the former type. Except for explanted cell elements, no cells appeared *in vitro* that could be identified as rudimental or transitional forms of glial or neuronal cells.

Electron Microscopy

Except for marginal differences the tumours showed a uniform morphological pattern. They will therefore be described together.

The histological structure of the medulloblastomas was rather unique. The cells were round, polygonal, or elongated, and densely packed. There were regions in all tumours which had a composition consisting of cross-sectioned cell processes surrounded by a circle of cell nuclei (Fig. 1). These structures corresponded to pseudo-rosettes seen by light microscopy. The intercellular space varied in size.

Main Tumour Cells

The nuclei were round, elliptic or polygonal. Occasionally they had invaginations or were even lobulated (Fig.2). In some areas two separated nucleus profiles within one cell were found. They might either be the cross-section of two separated nuclei or one bent nucleus, which had been cut twice (Fig.3). When well fixed

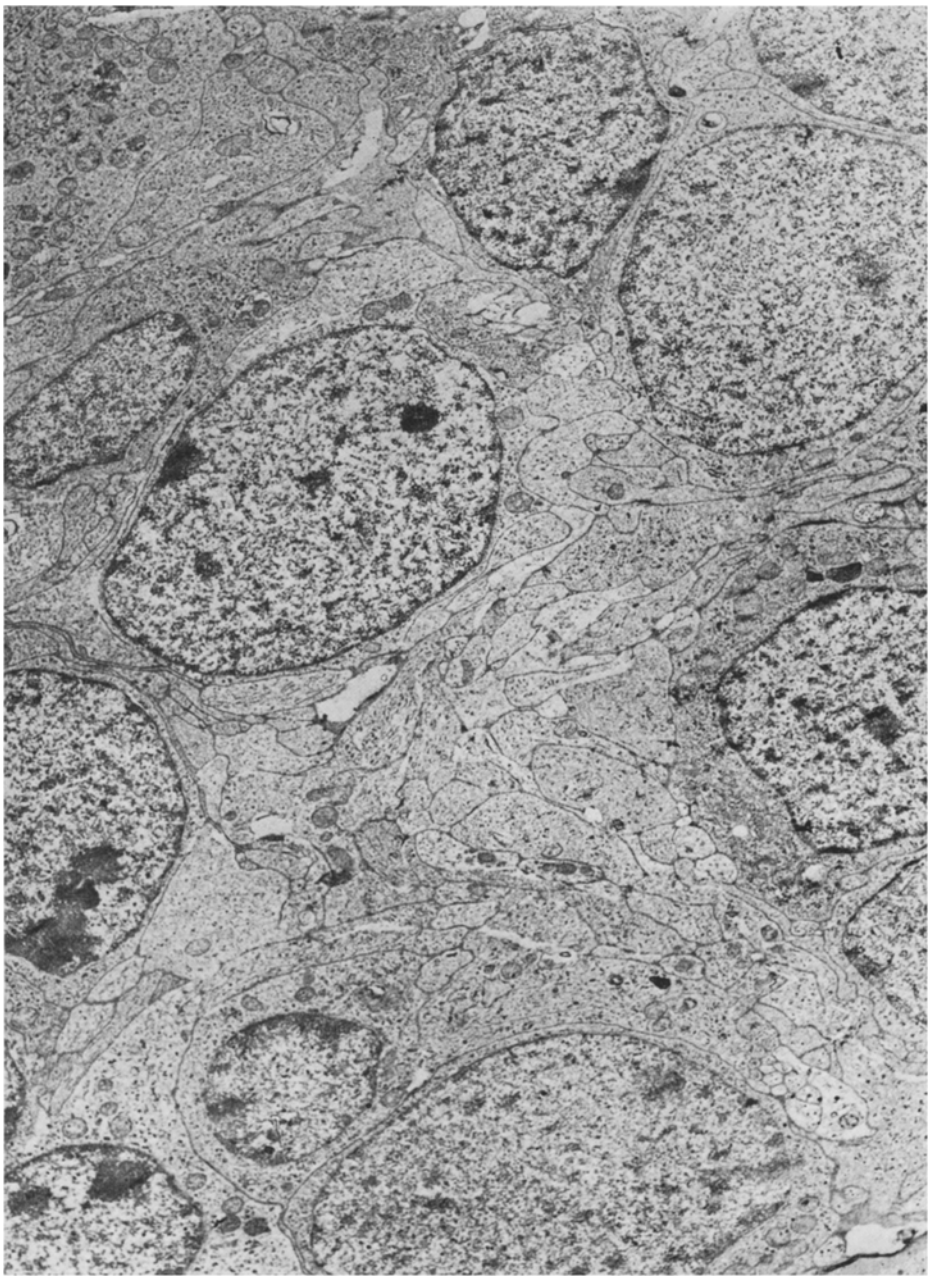

Fig. 1. Tumour cells lying in a circle, forming a pseudo-rosette. In the centre of it there are many cytoplasmic processes in cross-section

the caryoplasm was finely granulated and formed a few condensated zones, mostly located in the periphery (Fig. 2). Nucleoli were rare and seldom large. The perinuclear cytoplasm was usually a small rim around the nucleus containing only a few organells (Fig. 1, 2, 3).

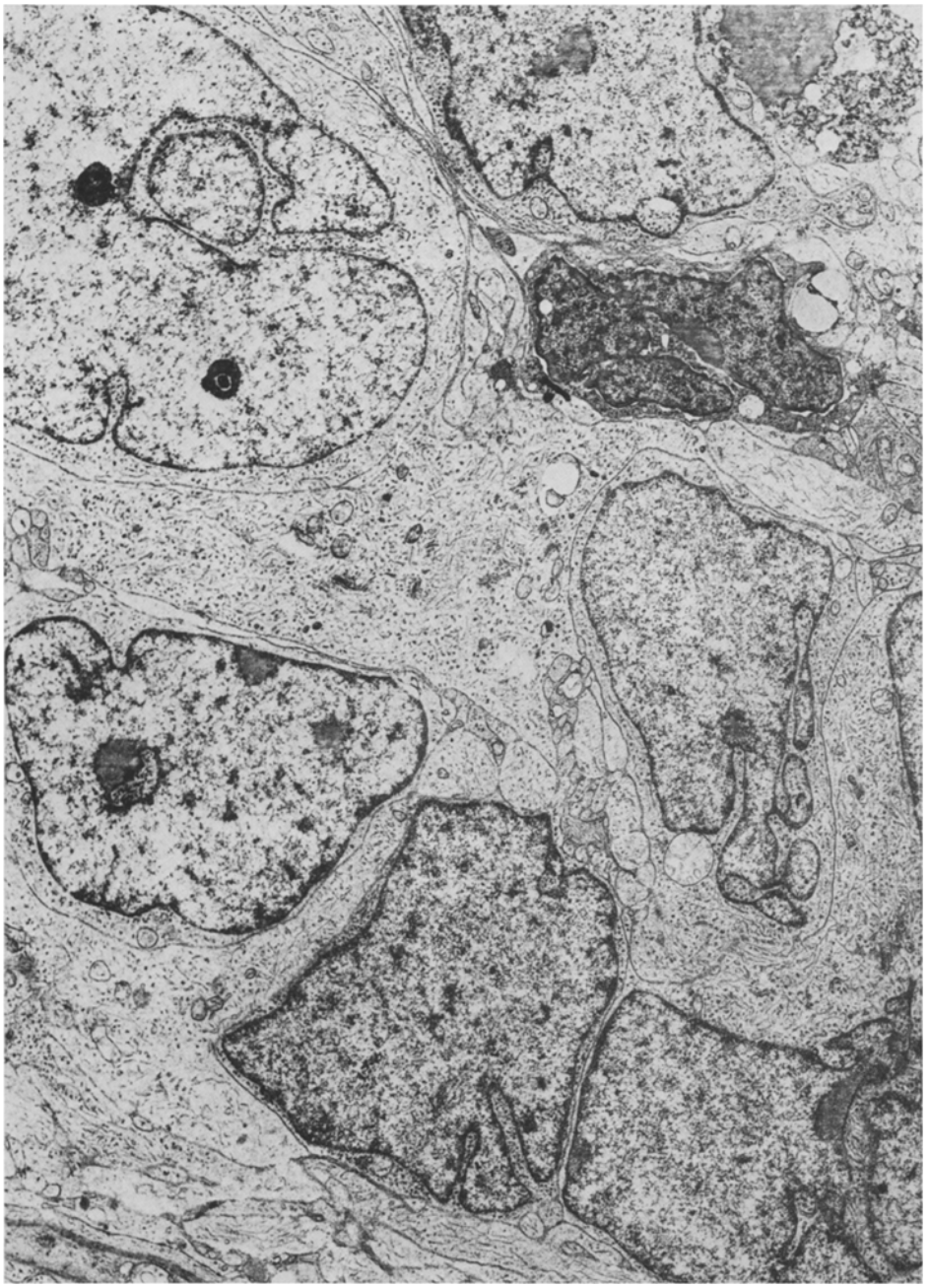

Fig. 2. The nuclei of these tumour cells are highly lobulated. Within the cytoplasm are irregularly running citofilaments and vesicles of the endoplasmic reticulum. One of the cells is degenerated. Its caryoplasm is darker than usual, the cytoplasm is only a small rim

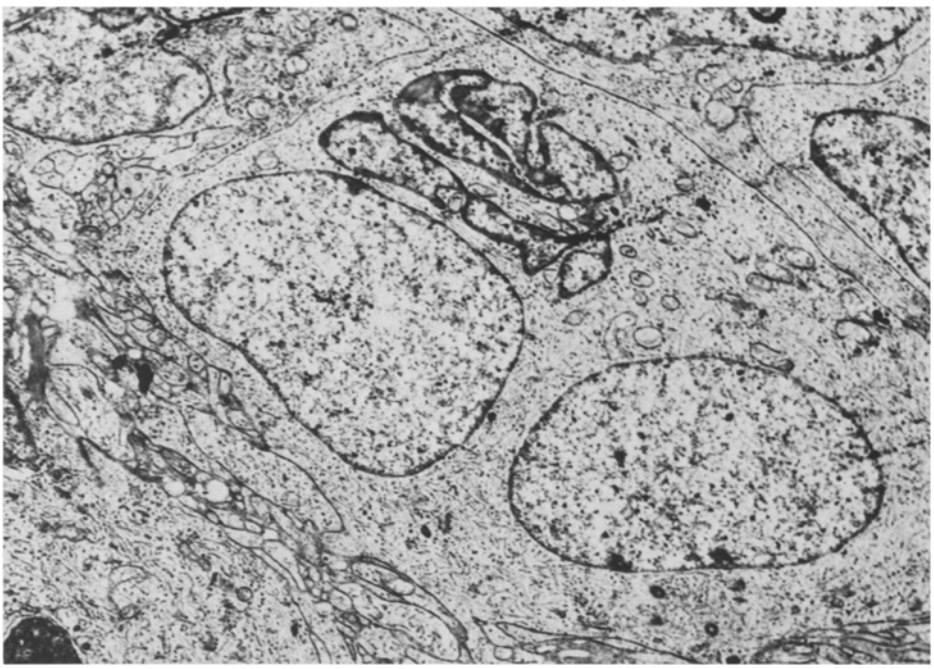

Fig. 3. A tumour cell, probably with 3 nuclei. At the left side some neuropil structures

Golgi zones were present in great amounts. Microtubules were constantly found and most obvious in more extensive cytoplasmic areas. In cytoplasmic processes they ran parallel to the axis of the process. In one case they dominated over all other organells. This tumour was further marked by the abundant cytoplasm of its cells which contained a lot of agranular endoplasmic reticulum (Fig. 4). It could not be excluded, however, that the tubules appearing as endoplasmic reticulum were only very strongly impregnated microtubules. In all other cases the endoplasmic reticulum was only poorly developed and rough-surfaced. Occasionally it was swollen by a homogenous mass of medium electron density. The mitochondria were small and few. The Golgi apparatus was usually located near the nucleus and consisted of swollen tubules and vesicles. A centriole was observed quite frequently. Three times we saw rudimental, intracellularly located and enveloped cilia and twice a diplosome. Cytosomes and glycogen granules were seldom observed.

The plasma membrane of the round or polygonal tumour cells was rather smooth. Cytoplasmic processes were infrequent and short in appearance (Fig. 5). In some cases their diameter became larger at a greater distance from their origin. The elongated cells had long processes that interdigitated occasionally.

Fat Cells

Light microscopy examination revealed fat in 6 tumours. In 4 of them fat could be seen also with the electron microscope. The fat was stored in cells in the form of more or less round, empty appearing holes without any membranous

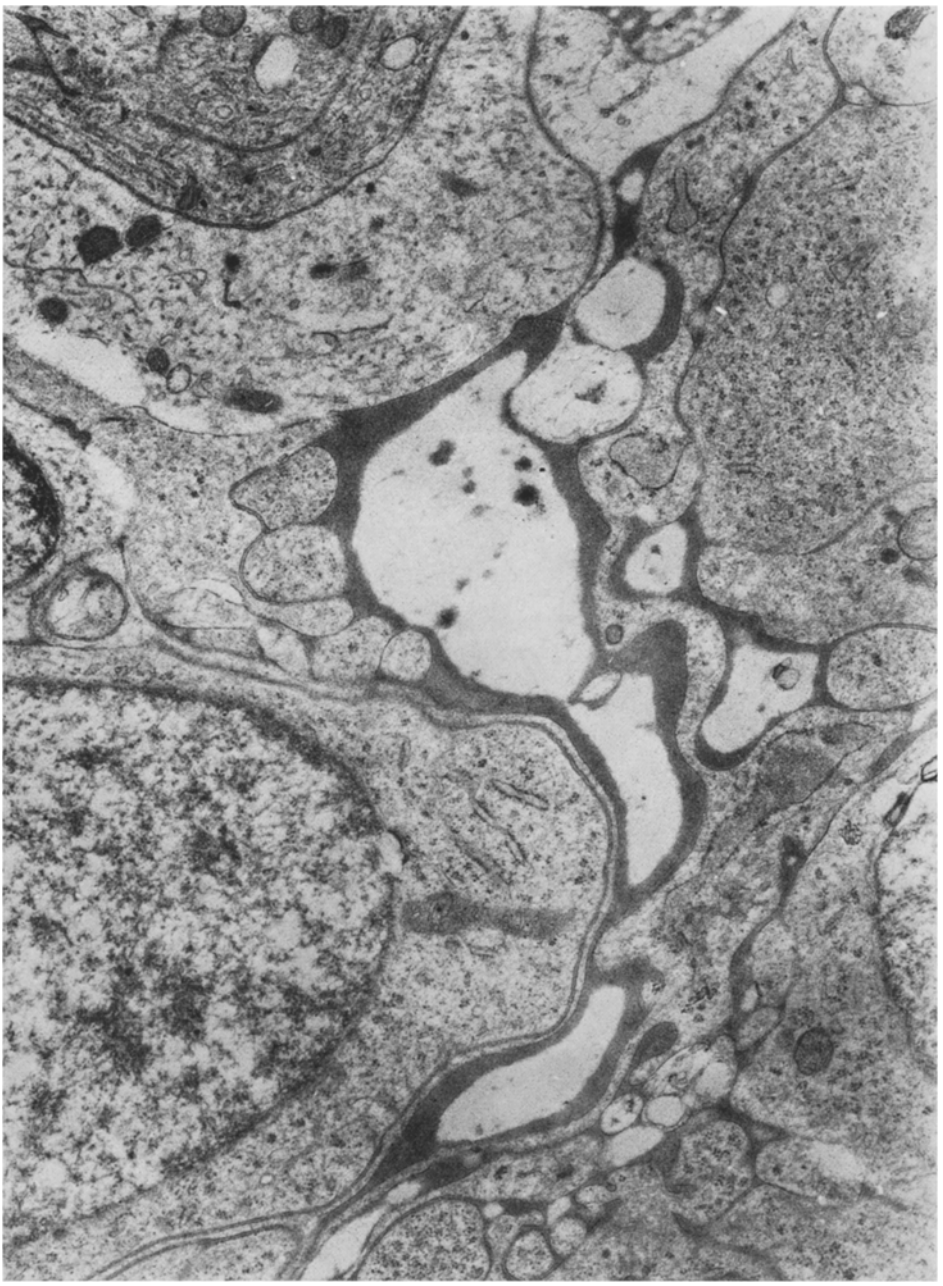

Fig. 4. Parts of tumour cells with many free ribosomes and a well developed endoplasmic reticulum. The intercellular space is wide and partly filled with a homogenous material

border. These fat droplets had a diameter of about 1 μ and confluated only in rare cases. While in 2 tumours some of the cells were completely deformed by accumulated fat, fat droplets were usually found in circumscribed cytoplasmic

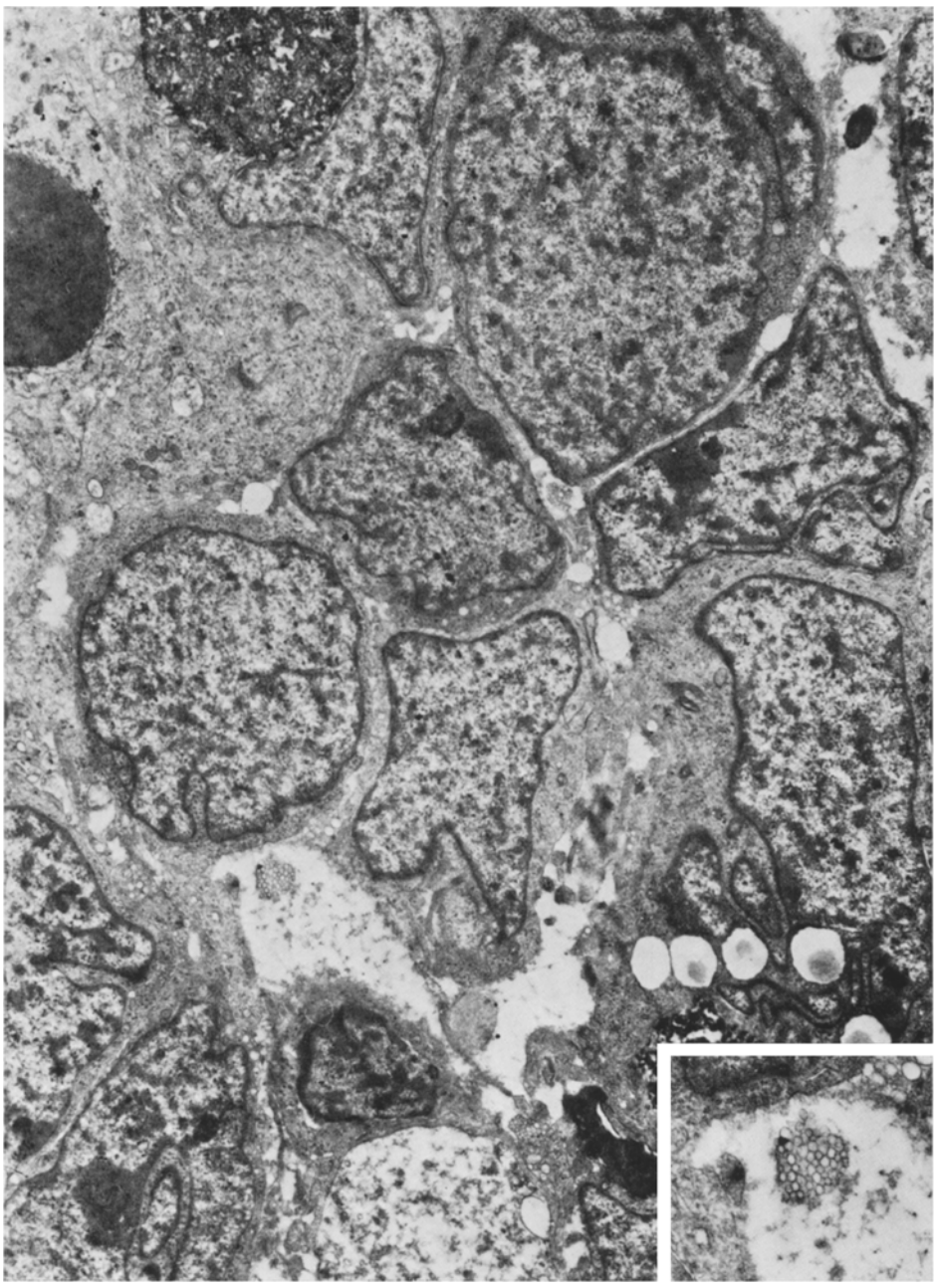

Fig. 5. Tumour cells that have only few cytoplasmic organells. At the bottom of the picture one sees fat droplets within the cytoplasm. The section shows vesical clusters magnified to a larger extent

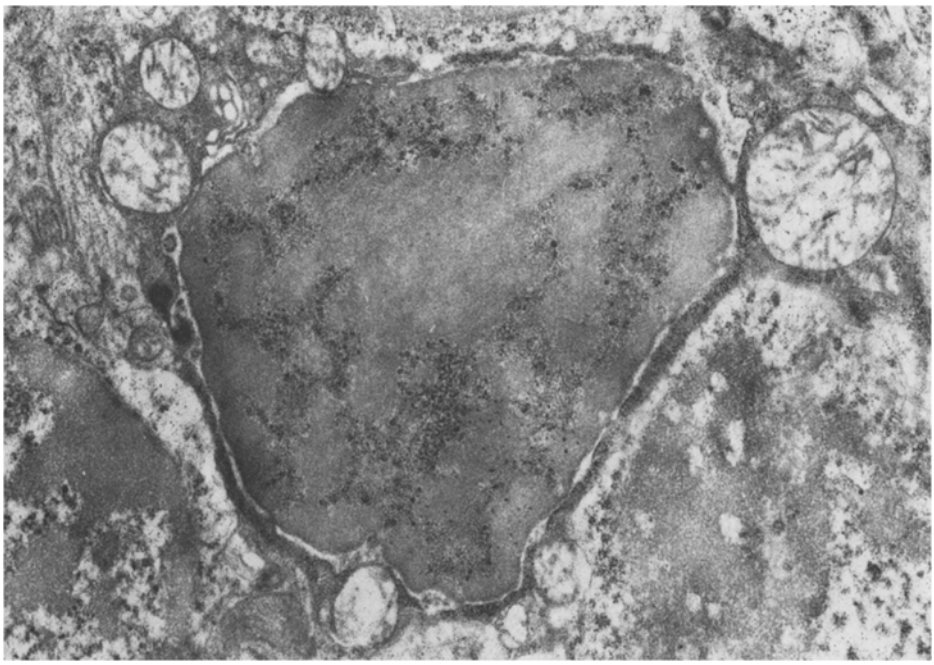

Fig. 6. A dark cell with a finely granulated caryoplasm. The nucleus membrane is intact. The very small cytoplasm contains more mitochondria than usual. They are extremely swollen

areas of a few cells (Fig.5). Transitional forms of main tumour cells and fat storing cells were observed in both cases.

Other cells contained smaller vesicles that were crowded at the periphery of the cytoplasm and enveloped by a membrane (Fig.5). They, too, were empty in appearance and had a diameter of about 150—500 nm. The vesicles, after being stained with azane and acridin orange, could be made visible in semi-thin sections.

Dark Cells

In 2 medulloblastomas a few very dark cells were observed. (Fig. 6). The chromatin of their nucleus was so finely granulated that it seemed to be almost homogenous, only regionally showing larger granules. The nucleus membrane was normal. The cytoplasm was only a small rim around the nucleus generally not wider than 150 nm and nearly as dark as the nucleus. It grew wider at one side of the cell where it contained numerous swollen mitochondria with a light matrix. These dark cells were sometimes found within regular tumour areas but more often were seen near cerebellar tissues.

Interstitial Space and Vessels

In some areas of the tumours the intercellular space, usually only a small cleft, grew considerably wider and was filled with a fine granulated material (Fig. 4). This material had some resemblance to a basement membrane but differed from it by its greater electron density and the absence of any stratification. In 3 tumours

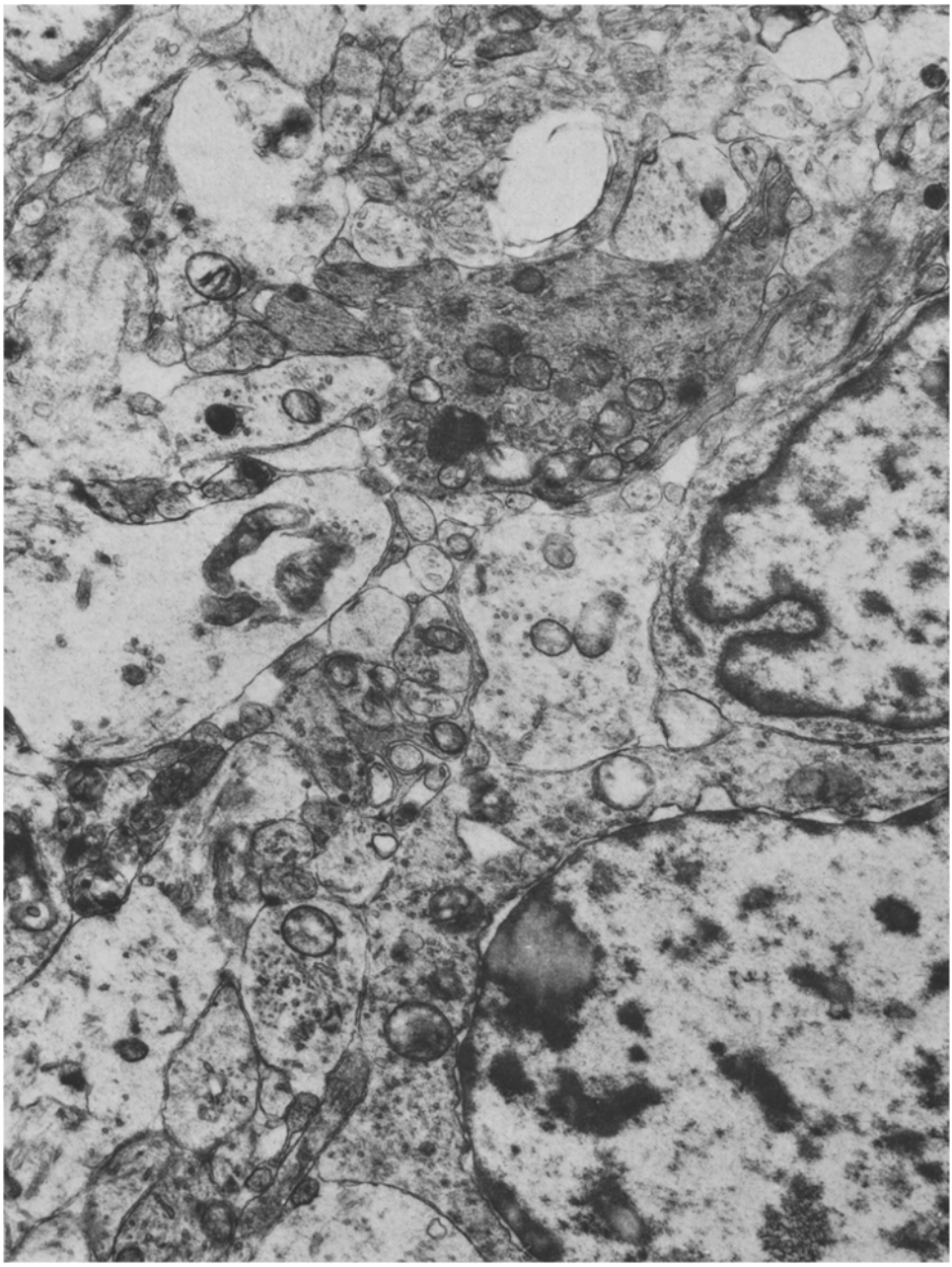

Fig. 7. Infiltration zone. Several cytoplasmic processes of uncertain nature. At the right tumour cells

the intercellular space contained many collagen fibres confined to a circumscribed area usually connected with the perivascular space. In 2 tumours the interstitial space was wide but without any contents. — The aforementioned vesical clusters were also found in the intercellular space (Fig.5).

In most cases it was impossible to decide whether a vessel was produced by the tumour or not. In cases where the perivascular astrocytic processes and a double basement membrane were still present the vessel was assumed to be pre-existent. But even when the capillaries were surrounded by tumour cells and only by one endothelial basement membrane, it could not definitely be concluded that this vessel was built by the tumour. The endothelial cells were sometimes swollen and showed in other cases a very active pinocytosis.

Cerebellar Tissue

The most frequent non-tumourous structures within the examined tissue parts were astrocytes. They generally showed reactive changes and a strong development of their cytofilaments (Fig. 7). They usually were connected with vessels.

Non-myelinated nerve fibres were difficult to distinguish from cytoplasmic processes of tumour cells, because sometimes they contained many microtubules. In cases where these cell processes contained axoplasmic vesicles or were arranged in patterns strongly resembling the neuropil they must be classified as nerve cell processes.

Discussion

Not all histological features of medulloblastomas observed by light microscopy can be analysed by the electron microscope because of the different dimensions of the two methods. However, the ultrastructure of some details helps us to understand better the architectural essentials of the tumour under discussion.

Observation showed that "pseudo-rosettes" are accumulations of cytoplasmic processes irregularly surrounded by cell nuclei. This has already been reported by Waga (1965) and confirmed early findings (Bailey and Cushing, 1925). Depending on the tissue infiltrated by the tumour the tumour cells form different patterns (Waga, 1965). Within the white substance the single cells arrange themselves according to pre-existing tissue structures and have an almost spindle-like form preserved by them even when the white substance has been destroyed. In other regions the cells are round or polygonal and tightly packed, thus corresponding to mosaic-like tumour aspects seen light microscopically. On the ground of these observation, however, it cannot definitly be excluded that it is a specific "inborn" tendency and consequently a form of differentiation of the medulloblastoma cells to arrange themselves in pseudo-rosettes around bundled cell processes, as the granular cells of the cerebellum do and is observed in neuroblastomas (Beltran, Leiderman, Stuckey, Ferrans, and Mogabgab, 1969). The characteristics of the tumour stem cells may well become much more manifest in the cyto-architecture than in cytological differentiations of the tumour.

It is of special interest that no reticulin could be demonstrated electron microscopically either in the form of microfibrilles or in that of collagen fibrilles although "reticulin" could be demonstrated by silver impregnation methods. The ultrastructural substrate of reticulin in medulloblastomas is a very osmiophilic, finely granulated substance with could be observed within the extracellular space in 5 of the tumours. This confirms the doubts concerning the specific nature of reticulin impregnation methods (Cervos-Navarro and Vazquez, 1969).

Our investigations confirm that medulloblastomas are constructed by a unique cell type from which all described cell types originate. Also the fat storage cells

show that they are related to the tumour cells. However, the dark cells may be an exception. As they are mostly neighbouring on the granular cells, it seems justified to regard them as necrobiotic granular cells. Dark cells with a more or less similar appearance as observed by us are quite common in reticulum sarcomas (Weinberger and Banfield, 1965; Horvat, Pena, and Fisher, 1969). This, on the other side, makes it more probable that they originate from tumour cells. In any case, the dark cells are degenerated cell elements as is proved by their swollen mitochondria. They are probably identical with the lymphocyte-like cells of medulloblastomas as described by Roussy and Oberling (1931), Schiffer, Fabiani, Monticone, and Cognazzo (1966). The two cell types that Sawada (1967) observed in medulloblastomas do not correspond to the types of cells observed by us. It is questionable whether all of the cells described by Sawada were really medulloblastoma cells. In contrast to most of the other authors (Raimondi, Mullan, and Evans, 1962; Waga, 1965; Raimondi, 1966; Escourolle and Poirier, 1967; Sawada, 1967) we could not find any indication for a neuronal or glial differentiation of the tumour cells, even if abortive.

The granular cells of the cerebellum have roughly the same size and form as the tumour cells. They too have only a small perikaryon, few organells, show only occasionally cytoplasmic processes and form glomeruli. In contrast to the tumour they are generally in contact with myelin sheaths orneuropil. They differ also in their chromatin structure which is composed of larger granules and is usually concentrated in condensation zones. Although this chromatin condensation is an artefact, it is characteristic for the granular cells of the cerebellum to form this artefact to a greater extent than many other cells (Dahl, Olsen, and Birch-Andersen, 1962; Herndorn, 1964). Goldin (1968), however, using only osmium tetroxide as fixative could not observe this chromatin condensation which is quite usual in undifferentiated embryonic cells.

The oligodendroglia and myelin sheaths are rapidly destroyed by the tumour because they depend to a high degree on the integrity of the tissue they constitute. Axons and astrocytes are preserved longer. The former because they may continue to be nourished by a cell body which is out of the tumour region, the latter because of their contact with intact capillaries. This is supported by the fact that in almost all tumourous areas the perivascular glia persisted and that the astrocytes preserved some capacity of reaction within the tumour.

Pre-existing astrocytes are commonly found in medulloblastomas (Gullotta, 1967). Some authors claim that these astrocytes are differentiations of tumour cells (Raimondi, Mullan, and Evans, 1962; Escourolle and Poirier, 1967). But Voigt (1968), who had carefully selected the tissue particles he investigated electron microscopically, could not find definite astrocytic tumour cells in medulloblastomas.

The small cytoplasmic rim containing only a few organells, except for ribosomes that may be present in greater numbers, is characteristic for medulloblastoma cells. They have this in common with undifferentiated stem cells of neural, glial or ependymal origin. In spite of this, the morphological similarity between medulloblastomas and embryonic tissue of the central nervous system cannot be a basis for histogenetic conclusions. The specific features of medulloblastoma cells are essentially the same as those of all embryonic tissues and can

therefore be interpreted at best as a hint for accelerated growth. The microtubules which Escourolle and Poirier (1967) have regarded as an indication for the neuroblastic origin of medulloblastomas are also found in embryonic cells of mesenchymal nature. A comparison with the fine structure of other cerebral tumours may help to define further characteristic features of medulloblastomas.

Gangliocytomas, although rather seldom, have been frequently investigated (Robertson, Hendry, and Vogel, 1964; Lee and Glasauer, 1968). They consist of large cells which have a broad cytoplasm and form synaptosomas or at least clusters of axoplasmic vesicles in cell processes. Sympathoblastomas and neuroblastomas similar to gangliocytomas are often further characterised by pheochromic granules (Luse, 1964; Gonzalez-Angulo, Reyes, and Navarette Reyna, 1965; Misugi, Misugi, and Newton, 1968). The cells of fibrillary and protoplasmic astrocytomas as well as of spongioblastomas contain masses of cytofilaments which are only occasionally polarized. This makes them easy to identify and to distinguish from medulloblastomas (Luse, 1962; Raimondi, 1966; Zülch and Wechsler, 1968). There is a similar difference from glioblastomas which, apart from the cytofilaments, differ by their pleomorphic form which is electron microscopically evident even in cases where the tumour observed light microscopically seems to be isomorphic. Oligodendrogliomas contain an abundance of mitochondria but few ribosomes (Luse, 1962; Raimondi, 1966; Zülch and Wechsler, 1968). They can therefore be easily distinguished from medulloblastomas although they often form cell arrangements similar to the latter. There are no similarities to the ultrastructure of meningiomas (Kepes, 1961; Gusek, 1962; Napolitano, Kyle, and Fisher, 1963; Castaigne, Escourolle, and Poirier, 1966; Rascol, 1966; Cervos-Navarro and Vasquez, 1969).

The fine structure of arachniod sarcomas, however, is essentially the same as that of medulloblastomas (Ramsey and Kernohan, 1964; Waga, 1965), as we confirmed by own studies. Already Rubinstein and Northfield (1964) have claimed from light microscopical studies that the two are only variants of one tumour type. The cells of arachnoid sarcomas are uniform, have many long cytoplasmic processes embracing the perikaryon, many free ribosomes, and a few mitochondria. The endoplasmic reticulum is often filled with an amorphous mass.

There are some ultrastructural similarities between sarcomas and medulloblastomas. However, it is the absence of any differentiation that most of them, e. g. Ewing sarcomas and reticulum cell sarcomas (Matusi, 1964; Weinberger and Banfield, 1965; Friedman and Gold, 1968), have in common with medulloblastomas. The same holds true for the reticulum cell sarcoma of the brain (Blackbourne and Waggener, 1967; Horvat, Pena, and Fisher, 1969).

Retinoblastomas were long thought to be neuronal tumours and closely related to medulloblastomas (Bailey and Cushing, 1925; Roussy, Oberling, and Raileanu, 1931; Zülch, 1968). But even ultrastructural investigations (Allen, Latta and Straatsma, 1962; François, Hansses and Lagasse, 1965; Ikui, Tominaga, Konomi, and Ueno, 1966; Fujikura, Kawai and Nukui, 1969) could not remove all doubts concerning this theory. Since Ts'o, Fine and Zimmerman (1969), however, showed that the Flexner-Wintersteiner rosettes of retinoblastomas represent an abortive attempt to form photoreceptor cells the neuronal origin of retinoblastomas was proved. The terminal bars of luminal limiting membranes, cilia with a $9 + 0$ pattern etc., are specific retinal characteristics of Flexner-Wintersteiner rosettes. The observations of Popoff and Ellsworth (1969) that retinoblastomas show nuclear alterations identical to those of the developing human retina were another although less strong support of this concept. In medulloblastomas no

specific cellular differentiations that would verify the theory of a neuronal origin of this tumour could be observed so far. Until now there are no observations justifying to regard medulloblastomas and retinoblastomas as related tumours.

References

Allan, R. A., Latta, H., Straatsma, B. R.: Retinoblastoma. A study of two cases by electron microscopy. Invest. Ophthal. **1**, 728—744 (1962).

Bailey, P., Cushing, H.: Medulloblastoma cerebelli. A common type of midcerebellar glioma of childhood. Arch. Neurol. Psychiat. (Chic.) **14**, 192—223 (1925).

Beltran, G., Leiderman, E., Stuckey, W. J., Ferrans, V. J., Mogabgab, W. J.: Metastatic ganglioneuroblastoma. Ultrastructural, histochemical, and virological studies in a case. Cancer **24**, 552—559 (1969).

Blackbourne, B. D., Waggener, J. D.: Ultrastructure of a primary cerebral reticuloendothelial sarcoma (microgliomatosis). J. Neuropath. exp. Neurol. **26**, 141 (1967).

Castaigne, P., Escourolle, R., Poirier, H.: L'ultrastructure des méningiomes. Etude de 4 cas en microscopie électronique. Rev. neurol. **114**, 249—261 (1966).

Cervós-Navarro, J., Vazquez, J. J.: An electron microscopic study of meningiomas. Acta neuropath. (Berl.) **13**, 301—323 (1969).

Dahl, V., Olsen, S., Birch-Andersen, A.: The fine structure of the granular layer in the human cerebellar cortex. Acta neurol. scand. **38**, 81—97 (1962).

Escourolle, R., Poirier, J.: L'ultrastructure du médulloblastome cérébelleux. Ann. Anat. path. **12**, 121—136 (1967).

François, J., Hansses, M., Lagasse, A.: The ultrastructure of retinoblastoma. Opthalmologica (Basel) **149**, 53—67 (1965).

Friedman, B., Gold, H.: Ultrastructure of Ewing's sarcoma of bone. Cancer **22**, 307—322 (1968).

Goldin, L. C.: The ultrastructural organization of the cerebellar cortex of the white rat. Zitologija (Moskow) **10**, 675—683 (1968).

Gonzalez-Angulo, A., Reyes, H. A., Navarette Reyna, A.: The ultrastructure of ganglioneuroblastoma. Observations on neoplastic ganglion cells. Neurology (Minneap.) **15**, 242—252 (1965).

Gullotta, F.: Das sogenannte Medulloblastom. Berlin-Heidelberg-New York: Springer 1967.

Gusek, W.: Submikroskopische Untersuchungen als Beitrag zur Struktur und Onkologie der „Meningiome". Beitr. path. Anat. **127**, 274—326 (1962).

Herndorn, R. M.: The fine structure of the rat cerebellum. II. The stellate neurons, granule cells and glia. J. Cell Biol. **23**, 277—293 (1964).

Horvat, B., Pena, C., Fisher, E. R.: Primary reticulum cell sarcoma (microglioma) of brain. Arch. Path. **87**, 609—616 (1969).

Ikui, H., Tominaga, Y., Konomi, I., Ueno, K.: Electron microscopic studies on the histogenesis of retinoblastomas. Jap. J. Ophthal. **10**, 88—96 (1966).

Kepes, J.: Electron microscopic studies of meningiomas. Amer. J. Path. **39**, 499—510 (1961).

Kersting, G.: Die Gewebezüchtung menschlicher Hirngeschwülste. Berlin-Göttingen-Heidelberg: Springer 1961.

Lee, J. C., Glasauer, F. E.: Ganglioglioma: Light and electron microscopic study. Neurochirurgica **11**, 160—170 (1968).

Luse, S. A.: Electron Microscopy of brain tumors. In: W. S. Fields and P. C. Sharkey: The Biology and Treatment of Intracranial Tumors. pp. 74—102. Springfield, Ill.: Ch. C. Thomas 1962.

— Synaptic structures occurring in a neuroblastoma. Arch. Neurol. (Chic.) **11**, 185—190 (1964).

Matakas, F.: Färbemethoden und histochemische Reaktionen an Kunststoffdünnschnitten für lichtmikroskopische Untersuchungen. Mikroskopie **23**, 337—344 (1968).

Matsui, K.: Ultrastructure of reticulosarcoma. Fourth Intern. Symp. on Reticuloendothelial System, pp. 36—37. Otsu-Kyoto 1964.

Misugi, K., Misugi, N., Newton, W. A.: Fine structural study of neuroblastoma, ganglioneuroblastoma, and pheochromocytoma. Arch. Path. **86**, 160—170 (1968).

Napolitano, L., Kyle, R., Fisher, E. R.: Ultrastructure of meningiomas and the derivation and nature of their cellular components. Cancer **17**, 233—241 (1963).

Popoff, N., Ellsworth, R. M.: The fine structure of nuclear alterations in retinoblastoma and in the developing human retina: in vivo and in vitro observations. J. Ultrastruct. Res. **29**, 535—549 (1969).

Raimondi, A. J.: Ultrastructure and the biology of human brain tumors. Basel-New York: S. Karger 1966.

— Mullan, S., Evans, J. P.: Human brain tumors: an electron microscopic study. J. Neurosurg. **19**, 731—753 (1962).

Ramsey, H. J., Kernohan, J. W.: Circumscribed sarcoma of the cerebellum. An electron microscopic study. J. Neuropath. exp. Neurol. **23**, 706—718 (1964).

Rascol, M.: Microscopie électronique de tumeurs de la méninge. Paris: Arnette 1966.

Robertson, D. M., Hendry, W. S., Vogel, F. S.: Central ganglioneuroma: A case study using electron microscopy. J. Neuropath. exp. Neurol. **23**, 692—705 (1964).

Roussy, G., Oberling, Ch., Raileanu, C.: Les neurospongiomes. Presse méd. **39**, 977—981 (1931).

Rubinstein, L. J., Northfield, D. W. C.: The medulloblastoma and the so-called "Arachnoidal Cerebellar Sarcoma". Brain **87**, 379—412 (1964).

Sato, K., Fujikura, T., Kawai, S., Nukui, H.: Retinoblastoma with intracranial extension. An autopsy case with light and electron microscope. Acta path. jap. **19**, 103—109 (1969).

Sawada, T.: The fine structure of gliomas. Acta med. (Fukuoka) **37**, 47—76 (1967).

Schiffer, D., Fabiani, A., Monticone, G. F., Cognazzo, A.: On the nature of the lymphocyte-like cells of medulloblastomas. Acta neuropath. (Berl.) **6**, 290—297 (1966).

Ts'o, M. O., Fine, B. S., Zimmerman, L. E.: The Flexner-Wintersteiner rosettes in retinoblastoma. Arch. Path. 88, 664—671 (1969).

Voigt, W.-H.: Elektronenmikroskopische Beobachtungen an menschlichen Medulloblastomen. Dtsch. Z. Nervenheilk. **192**, 290—309 (1968).

Waga, S.: A histological and electron microscopic study on medulloblastomas and cerebellar sarcomas. Arch. Jap. Chir. **34**, 436—454 (1965).

Weinberger, M. A., Banfield, W. G.: Fine structure of a transplantable reticulum cell sarcoma. I. Light and electron microscopy of viable and necrotic tumor cells. J. nat. Cancer Inst. **34**, 459—479 (1965).

Zülch, K.-J., Wechsler, W.: Pathology and classification of gliomas. In: Progress in Neurological Surgery, vol. 2, pp. 1—74, Ed.: H. Krayenbühl and P. E. Maspes. Basel: S. Karger 1968.

Prof. Dr. J. Cervós-Navarro
Institut für Neuropathologie
im Klinikum Steglitz
der Freien Universität Berlin
D-1000 Berlin 45, Hindenburgdamm 30