Lucien J. Rubinstein: enduring contributions to neuro-oncology

MELIKE MUT, M.D., M. BEATRIZ S. LOPES, M.D., AND MARK SHAFFREY, M.D.

Departments of Neurological Surgery and Pathology (Division of Neuropathology), University of Virginia Health System, Charlottesville, Virginia

Dr. Lucien Rubinstein is best remembered for his significant contributions to the field of neuropathology, particularly in the classification of nervous system tumors. His accomplishments in basic neuro-oncology and in the formulation of diagnostic principles reflected a unique talent for synthesizing fundamental clinicopathological concepts based on skillful diagnostic investigation and a thorough understanding of neurobiology. Dr. Rubinstein was the leader in the establishment of cell cultures from central nervous system (CNS) tumors. He meticulously analyzed both light and electron microscopic features of CNS tumors, recorded his findings, and patiently drew sketches to be shared generously with his colleagues and students. As a pioneer in neuropathology, in his work Dr. Rubinstein set the foundation for many enduring concepts in neurosurgery, neuro-oncology, neurology, and basic tumor biology.

KEY WORDS • Lucien Rubinstein • neuropathology • neuro-oncology • history of neurosurgery

BIOGRAPHICAL SKETCH

Lucien Rubinstein was born in Antwerp, Belgium on October 15, 1924, but immigrated to England during the Second World War. After a year at Queen Mary College, he was admitted to the Medical College of London Hospital, from which he graduated as an M.D. in 1952. After his military service, he joined the staff of the Bernhard Baron Institute of Pathology at the Royal London Hospital and collaborated with Professor Dorothy Russell. After a sabbatical year in which his time was split between the University of Minnesota and the National Institutes of Health, he moved to the US permanently in 1961. After 3 years at the Montefiore Hospital in New York, in 1964 he accepted a professorship in the Neuropathology Department at Stanford University. Dr. Rubinstein became the director of the Division of Neuropathology at the University of Virginia in April 1981.

Rubinstein had a very productive life; his contributions and collaboration with Dr. Dorothy S. Russell led to the textbook *Pathology of Tumours of Nervous System*,⁸⁵ which was first published in 1959. This book has been the most authoritative and scholarly work in the field of neuro-oncology. As Urich⁹¹ states in Dr. Rubinstein's obituary, "...[the] completely rewritten fifth edition was entirely his

own; the distillation of his experience and wisdom, and will remain his scientific testament." Dr. Rubinstein wrote five editions of *Pathology of Tumours of the Nervous System*, the *Atlas on Tumors of the Central Nervous System*, 139 articles in the literature, 14 book chapters, more than 50 published papers presented at national and international meetings, and the translation of the *Manual of Basic Neuropathology* by Escourolle and Poirier. He had editorial responsibilities for *Neuropathology and Applied Neurobiology*, the *Journal of Neuropathology and Experimental Neurology*, *Acta Neuropathologica*, *Clinical Neuropathology*, *Journal of Neuro-oncology*, *Cancer*, and *Virchow's Archiv*. He was an advisor to the Commission on the Histopathological Classification and Nomenclature of Tumors of the World Health Organization.

His professional life was dedicated to understanding the cytogenesis and differentiation of CNS tumors. He was a great teacher and a mentor who participated in the education of more than 50 trainees and visiting scholars from nine different countries. Neuropathology was his life and it was a pleasure so much above and beyond being a duty that he put all his soul into it; that could be well appreciated by his own words:

But what I missed most, perhaps, [at the conference at which he gave the talk] was [the] unashamed and hedonistic view that we've all concealed and which I will whisper to you. It is that neuropathology is an exquisitely enjoyable pursuit. That anything that is fun to do must obviously be fun to teach and it is by communicating this emotional involvement in

Abbreviations used in this paper: CNS = central nervous system; GBM = glioblastoma multiforme; GFAP = glial fibrillary acidic protein; NSE = neuron-specific enolase.

which we are wrapped, Mr. Chairman, that we can best fulfill two of our imperatives, the obligation to pass on to others the body of knowledge with which we have been entrusted and the secret wish we all have replicate ourselves.¹⁴

Dr. Rubinstein shared his unrivaled experience in clinical neuropathology and the applications of his novel ideas and translational research in the field of neuro-oncology (Fig. 1). He was the most consulted neuropathologist in the world; he had more than 6500 consultation cases over a period of 28 years.⁹¹

Dr. Rubinstein had diverse interests in life, including classical music, literature, theater, and wine. He was a good companion. Although English was not his native language, he mastered it; his colleagues admired his ability to describe morphological features with his precise English. Dr. Rubinstein passed away in 1990 due to the complications of a basilar artery aneurysm. He was in peace when he died. Just before his death he expressed to Dr. VandenBerg, "I pine for nothing. My work is finished, the book is done, and we've got research projects going but this can be carried on."¹⁴

CONTRIBUTIONS

We would like to discuss in more detail the concepts that Dr. Rubinstein developed during his career. Rubinstein had an infinite curiosity to elucidate the mechanisms of tumorigenesis in CNS neoplasms; he and his wife, Dr. Mary M. Herman, established cell culture systems for most of the CNS tumors known at that time and observed the cellular

behavior of these lesions in vitro. In their most eminent studies they assessed the growth kinetics of gliomas and medulloblastomas in matrix culture. Rubinstein translated the experimental data into clinicopathological terms and developed an algorithm for the lineages of cellular origin for gliomas and other types of CNS tumors. He described the importance of stromal and neural cell interactions in mixed gliomas and sarcomas.

Dr. Rubinstein did pioneering work in the application of GFAP to neuro-oncology, and he also performed immuno-histochemical characterization of numerous CNS and peripheral nervous system tumors. Moreover, he was the first in the history of neuropathology to describe several tumor entities, including true polar spongioblastoma, ependymoblastoma, pleomorphic xanthoastrocytoma, and infantile desmoplastic ganglioglioma. He analyzed desmoplastic medulloblastomas, cerebral neuroblastomas, astrocytoma variants, pineal parenchymal tumors, ependymal neoplasms, and meningeal tumors to define a thorough description of those lesions in the scheme of classification of CNS tumors. Rubinstein relied on his own experience and research to postulate that medulloblastomas had a unique behavior and origin differentiating them from the rest of the primitive neuroectodermal tumors.

Dr. Rubinstein always correlated his neuropathological findings with clinical outcome, and he helped the clinicians by providing them with scholarly data about the prognosis and precise therapeutic approaches to a given tumor entity.¹ Among hundreds of examples, we may mention his extensive studies on the malignant evolution of primary brain tumors; he carefully reserved the term "anaplastic oligodendroglioma" as a diagnosis only in the patients with rapidly growing, highly cellular, and poorly differentiated oligodendrogliomas, relying on his longitudinal follow up of those patients. Also, he extensively studied the mechanism of extracranial metastasis of GBMs. Important contributions beyond the description of CNS tumors included characterization of the causative viruses of progressive multifocal leukoencephalopathy and subacute sclerosing panencephalitis, and definition of the complications of radiation therapy in the CNS.⁷⁶

Theories of Tumorigenesis in Nervous System Neoplasms

Dr. Rubinstein's principal interest was in tumor pathogenesis. He advanced the "neoplastic vulnerability" theory; he believed that neoplastic transformation resulted from the interaction of several factors: 1) the existence of a reserve population of stem cells; 2) the capability of differentiated cells to reenter the kinetic cycle; 3) the number of replicating cells at risk at a particular time; 4) the length of time during which a particular cell population remained in the cycle; 5) the state of current differentiation and the future differentiation potential of that population; and 6) the steps of differentiation that were achieved in successive cell generations. He thought that the incidence of different types of CNS tumors could be correlated with the width of the window of neoplastic vulnerability. He proposed the existence of a narrow window of vulnerability for rare tumors such as medulloepitheliomas, cerebral neuroblastomas, gangliogliomas, and ependymoblastomas. By contrast, he wrote of a relatively wider window for more common entities such as medulloblastomas, astrocytomas, mixed astrocytomas and

Fig. 1. Photograph of Dr. Rubinstein giving a lecture to his fellows at Stanford University in 1977.

oligodendrogliomas, and glioblastomas. With his theory, he tried to explain radiation-induced gliomas, in particular those arising after the apparently successful treatment of acute lymphocytic leukemia.⁶³

Rubinstein properly documented the vascular proliferation of GBMs in vitro and in animal models. He postulated that mitogenic factors were secreted by GBM cells to promote angiogenesis. He was one of the first authors to describe the coexistence of gliomatous and sarcomatous elements in CNS tumors (gliosarcomas and sarcogliomas) that will be explained later in the text.^{66,72}

Cell Cultures and Animal Models

One of Rubinstein's greatest achievements in neuro-oncology was the development of reproducible tumor cell cultures from a variety of CNS lesions. Rubinstein believed that the organ culture technique was a suitable system for the study of cell kinetics in human malignant gliomas and for analysis of the in vitro effects of chemotherapeutic agents. He meticulously documented the immunohistochemical staining characteristics of various types of tumors in vitro to understand their structural and biochemical qualities. Immunohistochemical staining was applied to identify the various stages of neuronal and glial cell maturation in experimental neuro-oncogenesis.^{32,49,62} He studied growth kinetics in the human medulloblastoma and glioblastoma cell lines in vitro,^{27,49} and he investigated rat C-6 glioma in animal models in vivo.²³ Moreover, cell cultures enabled him to comprehend the histogenesis of multinucleated giant cells in GBMs, the astrocytic nature of both the small cells and the giant multinucleated cells, and the capacity of both cell populations to synthesize DNA in culture and to demonstrate invasiveness.³⁰ VandenBerg and Rubinstein developed a mouse teratoma model to elucidate the various stages of differentiation in neuroepithelial stem cells, and with their colleagues they explained neural differentiation in human embryonal CNS tumors.^{19,25,58,93} Immunohistochemical findings of differences between intracranial germinomas and their gonadal equivalents indicated that in the former, early epithelial or mesenchymal differentiation of the primordial germ cells might be present. These findings drew attention to the heterogeneous cellular composition of these otherwise morphologically homogeneous-appearing tumors and, especially in the posterior fossa, to their transitional links to the immature teratomas.^{9,50}

Application of Immunohistochemistry

One of Dr. Rubinstein's major contributions to diagnostic neuropathology was improving the diagnostic accuracy in challenging cases by application of immunohistochemical testing. He was one of the first investigators in the field of neuropathology who routinely applied immunohistochemical stains for neural markers, GFAP, NSE, the three protein subunits of neurofilaments, myelin basic protein, S100 protein, and the Leu-7 (HNK-1) and non-neural markers (vimentin, desmin, cytokeratins, Factor VIII, alpha-fetoprotein, human chorionic gonadotropin, and immunoglobulins) for the characterization of CNS tumors.^{5,15,18,56,58,67}

Use of GFAP for Immunostaining

Rubinstein firmly established the recognition of GFAP expression as evidence of astroglial histogenesis and differentiation, and he established its applicability to the identification of tumors of astrocytic origin. He clearly demonstrated that GFAP was expressed in normal fibrillated astrocytes, and in reactive and neoplastic cells of astrocytic lineage. Its presence in nonastrocytic CNS tumors was related either to the development of gliofibrillogenesis (ependymomas), to ontogenetic factors (oligodendrogliomas, choroid plexus papillomas), or to uptake of the protein from adjacent reactive astrocytes (capillary hemangioblastomas). Using GFAP, Rubinstein and Kepes³⁶ described the pleomorphic xanthoastrocytoma as an astrocytic tumor, not one of meningotheial origin as others had postulated, and defined the GFAP expression in proliferating and invading cells in GBMs and medulloblastomas.³¹

Rubinstein tried to distinguish confusing phenomena of metastasis into gliomatous or epithelial differentiation of glial cells. Using immunohistochemical tools, he delicately distinguished aberrant epithelial neoplastic differentiation in a malignant glioma.⁴⁶⁻⁴⁸ He defined GFAP positivity in choroid plexus papillomas, stating that it is suggestive of a primitive neuroepithelial (ventricular) cell origin for these tumors, given the fact that normal choroid plexus epithelium lacks GFAP expression.⁷⁷

Rubinstein defined GFAP immunostaining characteristics in GBM. He showed that the percentage of GFAP-positive cells was inversely related to the anaplastic character of the tumor. He used GFAP to define atypical variants of GBMs in which diagnostic difficulties were encountered. He was interested in a rare type of this lesion, namely a GBM with heavily lipidized (foamy) tumor cells that generally obscured their glial nature and distinguished them from the pleomorphic xanthoastrocytomas.³⁸ The immunohistochemical characteristics of oligodendrogliomas were enigmatic for him and he tried to characterize them with the markers available at that time, describing these markers' usefulness in oligodendrogliomas.^{15,51,55}

Use of NSE as a Marker

Another marker of diagnostic importance frequently used by Rubinstein was NSE. He studied the cellular distribution and intracellular localization of NSE by immunoelectron microscopic findings in neurons of the cerebrum, cerebellum, and brainstem in rats and mice.⁹⁸ He demonstrated differential expression of NSE in neural and non-neural tumor cell lines of rat and human origin, and interestingly, he found the expression of NSE in reactive astrocytes, schwannoma cells, and gliomas, in addition to neurons and neuroendocrine cells. He hypothesized that glial cells containing only the nonneuronal form of enolase might transform into neoplastic cells. When the transformation took place, the elevated metabolic demands imposed on neoplastic and reactive glial cells (and on some extraneural tumors) necessitated the opening up of metabolic pathways that were normally operative only in neurons and neuroendocrine cells, therefore resulting in the synthesis of the more stable neuron-specific form of enolase.^{96,97,99} He confirmed the positive reaction for NSE in a variety of glial and other cerebral neoplasms obtained intra-

operatively, including glioblastoma, astrocytoma, oligodendroglioma, ependymoma, medulloblastoma, pineocytoma, meningioma, and choroid plexus papilloma; peripheral neuronal tumors such as neuroblastoma, ganglioneuroma, and paraganglioma; and endocrine and neuroendocrine tumors. Because it was expressed in many cells as well as in reactive astrocytes, he ultimately considered NSE to be of less value in the differential diagnosis of CNS tumors.^{95,100}

Use of the Leu-7 Marker

The Leu-7 (HNK-1), a hematological marker, has been found to cross-react with cells of schwannian origin, and Rubinstein found it helpful in the differential diagnosis of schwannomas and neurofibromas from other soft-tissue neoplasms.⁵⁴ He used several markers for peripheral nerve sheath tumors. Normal, reactive, and neoplastic perineurial cells stained consistently for epithelial membrane antigen, whereas only Schwann cells expressed S100 in addition to Leu 7.⁵³ He suggested that human nerve sheath tumors contained cells with polypeptides that shared epitopes with GFAP; however, these polypeptides differed from astrocytic GFAP by at least one epitope.⁸⁹

Clinical Applications for Diagnostic Advances and Classification Schemes

Pleomorphic Xanthoastrocytomas. Rubinstein, Kepes, and Eng were the first authors to describe “pleomorphic xanthoastrocytoma (PXA)” as a distinct entity.³⁸ In their original description, pleomorphic xanthoastrocytoma was characterized as a tumor most often presenting superficially over the cerebral hemisphere of young patients, involving the leptomeninges extensively. These tumor cells displayed marked pleomorphism, including bizarre giant cells and a number of mitotic figures, but no necrosis. Many contained large amounts of lipid in their cytoplasm and were surrounded by reticulin fibers, thus simulating a mesenchymal tumor. Kepes, et al., suggested that subpial astrocytes could be the origin for this neoplasm. They established that pleomorphic xanthoastrocytoma was a glial neoplasm with GFAP positivity, and they provided the clinical evidence that described its transformation into a GBM on its recurrence.³⁷ In their series of 35 patients, most had a good prognosis, even in the absence of postoperative radiation treatment. Most of the recurrent tumors did not reveal a change in histological character; malignant evolution was exceptional. Kepes³⁶ reviewed the steps taken with Rubinstein that led to the recognition of the basic characteristics of this neoplasm and its designation as an independent entity.

Gangliogliomas and Ganglioneuromas. Rubinstein had a personal series of 120 ganglion cell tumors in which he studied the electron and light microscopic features, revealing three distinct tumor cell types. In addition to neuronal and astrocytic components, he clearly defined a mesenchymal component that could be responsible for the rich connective tissue stroma that was characteristic of the ganglion cell tumors in the CNS.⁸⁰ As he did for all for other types of CNS tumors, Rubinstein explained the histogenesis and mode of development for these lesions and correlated this with their clinical evolution. In his series of 120 patients, gangliocytomas and gangliogliomas had a good prognosis due to their slow growth, well-differentiated cel-

lular features, small size with good demarcation of the borders, and surgical resectability. His definition of malignant transformation of the astrocytic component is now widely accepted. He described the anaplastic transformation of gangliogliomas to GBMs and avoided the use of “ganglioneuroblastoma” for malignant forms of this lesion. He also confirmed the metastatic potential of this entity.

Rubinstein clearly demonstrated the association of congenital anomalies with gangliogliomas. He clarified the situation when tuberous sclerosis was associated with gangliogliomas. He stated that those gangliogliomas were distinct from tubers or subependymal giant cell astrocytomas with questionable neuronal components. His study with Bonnin, et al.,⁷ suggested that the subependymal giant cell astrocytomas, especially those associated with tuberous sclerosis, included cells that were apparently unable to express GFAP. Some of the tumor cells expressed the 68-kD neurofilament protein with incomplete expression of neuronal differentiation.

Astroblastomas. The definition and clinical characteristics of astroblastomas were described by Rubinstein and coworkers^{4,79} in a series of 23 patients. Two distinct histological types were encountered: low-grade and high-grade. Rubinstein performed a tissue culture and ultrastructural examination of cases and suggested a possible origin from tanycytes, a transitional cell between the primitive glioblast and the mature ependymal lining cell. Clinicopathological correlation was not well established because of the natural history of astroblastomas. Many authors now question the existence of this glioma and propose that such tumors are a subtype of astrocytomas. Nevertheless, astroblastomas are categorized under the heading of “glial tumors of uncertain origin” in the current World Health Organization classification.³⁹

Ependymomas/Ependymoblastomas. Rubinstein studied characteristics of ependymomas both ultrastructurally and in vitro.^{59,60} He demonstrated the neuroglial nature of ependymal tumors by immunohistochemical staining and in tissue cultures of myxopapillary ependymomas, depicting the potential of the ependymal cells to form glial fibrils. Nevertheless, he considered ependymoblastomas to be of embryonal origin.⁶⁵ He reviewed a series of ependymoblastomas and defined their clinicopathological aspects and histological characteristics, which distinguished them from anaplastic (malignant) ependymomas.⁴⁵ In his personal series of malignant ependymomas, there was no correlation between the tumor’s histological features, site, or likelihood of recurrence, and he pointed out the contrasts with the known correlations that exist in astrocytomas. This remarkable study by Ross and Rubinstein⁶¹ remains one of the best analyses of the lack of correlation between the histopathological features of ependymomas and the tumor’s clinical behavior.

Embryonal Central Neuroepithelial Tumors. Rubinstein was a strong opponent of the concept that all primitive neuroectodermal tumors could be classified into a single neuroepithelial entity.^{69,75,83} He made a lifelong effort dedicated to understanding the neurobiology of these fascinating tumors. He advocated that neoplastic transformation essentially involved replicating stem cells in tissues that can regenerate, and in the human brain such cells were found mostly in the course of CNS development. He suggested a

cytogenetic scheme to serve as a frame of reference for a classification of embryonal CNS tumors that would account for the different histological entities and for the range of and the restrictions on their differentiating capabilities. Among the embryonal CNS neoplasms; the cerebral medulloepithelioma, the cerebral and cerebellar neuroblastomas, the primitive polar spongioblastoma, and the ependymoblastoma showed characteristic morphological features and a correspondingly distinctive cellular differentiating potential. The differentiating capabilities of the cerebellar medulloblastoma, the pineoblastoma, and the retinoblastoma were also distinctive, and were determined by the cytogenesis of the area of the CNS in which the tumors originate. Rubinstein's observations, which still preserve their validity, revealed that Flexner–Wintersteiner rosettes were common in retinoblastomas, infrequent in pineoblastomas, rare in medulloblastomas, and absent in cerebral neuroblastomas. Calcification was common in retinoblastomas and cerebral neuroblastomas, but absent in medulloblastomas. Pale islands of tumor cells, denoting some degree of neuronal maturation, were present in childhood medulloblastomas, and only occasionally in medulloepithelioma. True ependymal rosettes were found in ependymoblastomas.^{45,65} Medullary rosettes and tubes defined the medulloepithelioma. Recent developments in molecular and genetic diagnostic methods support Rubinstein's theory that embryonal central neuroepithelial tumors are distinct from medulloblastoma (referred to later in the text).

Rubinstein had a major interest in medulloepithelioma, desmoplastic infantile ganglioglioma, pineoblastoma, and medulloblastoma. He defined them as “multipotential” in light of their capacity to undergo divergent differentiation.⁹³ Nevertheless, he concluded that even in very primitive neoplastic neuroepithelium, immunohistochemical evidence of early commitment of some of the cells to either a neuronal or glial lineage could be demonstrated.¹¹ He stated that the class III beta-tubulin isotype was a very early neuronal marker shown in cells with early neuronal commitment both in the normal neurocytogenesis and neoplastic processes in human embryonal tumors of the CNS. He defined the early stages of progressive neuroepithelial differentiation and neuronal commitment in primitive neuroepithelium in medulloepithelial rosettes.^{8,10} He associated those lesions with other congenital renal tumors to understand their pathophysiological features.⁶ The aforementioned studies conducted by Rubinstein and Vandenberg in a mouse teratoma model with neuroepithelial stem cells delineated a pathway by which human embryonal CNS tumors could differentiate into divergent lineages.

Cerebral Medulloepithelioma. The cerebral medulloepithelioma is a rare, embryonal, multipotential, central neuroepithelial neoplasm of childhood. The microscopic appearance of medulloepithelioma is highly distinctive, with pseudostratified columnar to cuboidal epithelium in a papillary or tubular pattern. Pale islands in the tumor were shown by Rubinstein to display the entire range of differentiation, from embryonal to mature cells of both glial and neuronal lineage (that is, primitive medullary epithelium, spongioblasts, astrocytes, oligodendroglia, ependymoblastic and ependymal cells, neuroblasts, and mature ganglion cells).^{11,86} The prognosis is poor, with a median survival of 6 months, and these tumors frequently cause

cerebrospinal seeding. Cerebral medulloepithelioma is rarely associated with other embryonal tumors.⁶

Medulloblastoma. The medulloblastoma is essentially a cerebellar neoplasm with various degrees of maturation, chromosomal abnormalities, and clinical characteristics. Rubinstein was fascinated by these tumors and firmly insisted on maintaining a separation between the cerebellar medulloblastoma and the other embryonal neoplasms found elsewhere in the CNS, on both cytogenetic and practical grounds.^{2,6,15,24,31,32,34,42,44,49,63,70,74,81,83,84,92} He supported the hypothesis that the external granular layer is the origin of the medulloblastomas. Along with Kadin and Nelson,³³ he reported the fetal granular layer adjacent to the tumor with marked neoplastic proliferation, which formed an irregular sawtooth pattern extending into the molecular layer in one patient with cerebellar medulloblastoma. A recently developed body of evidence based on molecular and genetic experimental studies delineated sonic hedgehog signaling as a contributor to medulloblastoma formation, and mutations of the sonic hedgehog receptor *PATCHED* are associated with medulloblastomas, which develop from the external granular layer.²²

Developmentally, the cerebellum has a unique pattern, with an external-to-internal migratory pathway and postnatal residuum of embryonal cells. Normally, granule cells arise from committed precursors that migrate during late embryonic development over the surface of the cerebellum to form a superficial germinal layer, the external granular layer.¹⁰¹ Perinatally, this layer expands dramatically as immature granule cell precursors in the outermost regions proliferate under the mitogenic influence of sonic hedgehog signaling.¹⁰² In the deeper region of the external granular layer, granule cells withdraw from the cell cycle and migrate inward, terminally differentiating in the internal granular cell layer. Unlike most neuronal populations, granule cell precursors remain mitotically active even after birth and constitute potential targets for transforming insults.¹⁰¹ Rubinstein established the concept of the differentiating bipotential of the cerebellar medulloblastoma by using experimental *in vitro* data. He developed an organ culture system from an undifferentiated human medulloblastoma and proved the ability of the tumor to differentiate into both astrocytes and neuroblasts *in vitro*.²⁴

Desmoplastic Medulloblastomas. In 1964, the term “desmoplastic” was first suggested by Rubinstein for the interpretation of fibrous connective tissue proliferation in the subarachnoid spaces due to tumor invasion.⁸⁴ Neoplastic cells in the reticulin-free pale islands were shown to have features of predominantly neuronal and, to a lesser degree, astroglial differentiation by Rubinstein (Katsetos, et al.³⁴). This concept of a more differentiated cell population in desmoplastic medulloblastoma has been recently confirmed, and appears to be the basis for the better prognosis and treatment response in this category of medulloblastomas. This concept has initiated a new proposed classification of medulloblastomas.¹⁷

Primary Cerebral Neuroblastoma. Horten and Rubinstein²⁸ reported the largest series of primary cerebral neuroblastomas and defined three subtypes: classic, transitional, and desmoplastic. The desmoplastic and the transitional forms were less likely to exhibit differentiation to mature ganglion cells, whereas the classic type was

similar to peripheral neuroblastomas and cerebellar medulloblastoma. Ganglionic differentiation was noted mostly in the classic variant. No correlation could be made in his series between survival times and histological variant of the tumor or the presence of ganglionic differentiation.^{2,28} He also described a pigmented olfactory neuroblastoma. The tumor contained a large amount of pigment, most of which had the histochemical reactions of melanin, but some of which had that of lipofuscin. The pigment was interpreted as presumably representing a modified catecholamine degradation product.¹³

Rubinstein's series of 70 cases revealed a relatively better survival rate compared with other malignant central neuroepithelial tumors, with a 3-year survival in 60% of the patients. Nevertheless, recurrence was common and usually occurred within 3 years after surgical treatment. Whole craniospinal axis radiation was suggested because of the high rate of craniospinal seeding.

Desmoplastic Infantile Ganglioglioma. Biologically this entity, which has a more favorable prognosis, was concluded to be closer to gangliogliomas than to neuroblastomas.⁹⁴ VandenBerg and Rubinstein reclassified four of his previous cerebral neuroblastoma cases as "desmoplastic infantile gangliogliomas" with distinct pathological features, intense desmoplasia, and the frequent presence of divergent astrocytic and ganglionic differentiation. Despite the lesion's voluminous size, he suggested that after the successful complete or subtotal resection, these tumors followed a favorable clinical course similar to gangliogliomas rather than primitive cerebral neuroblastomas.

True Polar Spongioblastoma. Rubinstein described this rare embryonal neoplasm with primitive characteristics, in which the tumor cells were aligned in parallel and palisading fashion and separated by vascular stroma. He classified divergent lines of differentiation into astrocytoma or oligodendroglioma.⁶⁸ Rubinstein believed in the malignant potential of polar spongioblastomas as the transitional stage in the evolution of diffuse astrocytomas into GBMs, despite the fact that there were patients with long survival times in his series.

Pineal Lesions. While demonstrating divergent differentiation in pineal parenchymal tumors, Rubinstein tried to explain the origin, structure, and function of the pineal gland. He showed that the human pineal gland retained some of the cytochemical characteristics of photoreceptor cells recognized by the monoclonal antibody A9-C6, and that S-antigen immunoreactivity might occasionally be expressed in pineal parenchymal tumors.⁵⁷

Rubinstein explained the differentiation of benign symptomatic pineal cysts from pineocytomas.⁴⁰ In his series of pineoblastomas and pineocytomas he showed the potential of those tumors to differentiate along glial or ganglionic lines, or both, and defined the ultrastructure of one pineocytoma that had neuronal and astrocytic differentiation, which demonstrated the presence of numerous microtubules, clear-centered and dense-core vesicles, and synaptic complexes. He delineated the clinical behavior, therapeutic implications, and prognosis of pineocytomas according to the line of differentiation. He indicated that pineocytomas without cellular evidence of further differentiation were clinically malignant, but with a somewhat weaker tendency to metastasize than pineoblastomas; pin-

ocytomas with astrocytic differentiation might be either slowly growing or malignant; pineocytomas with neuronal or with neuronal and astrocytic differentiation were relatively benign. According to the conclusions reached from his observations, he suggested as a management strategy for pineal region tumors that radiation should be administered to the entire neuraxis for patients with pineoblastomas and malignant pineocytomas.^{26,64} Pineal tumors are a clear example of how he used his expertise and unique talent to correlate the neuropathological findings with the patients' prognosis.

Retinoblastomas. While showing positivity of neuron-associated class III beta-tubulin isotype (h beta 4), microtubule-associated protein 2, and synaptophysin in 26 retinoblastomas in situ and the human retinoblastoma cell line WERI-Rb1, Rubinstein proved early neuronal commitment with no evidence for a divergent (that is, neuronal and glial) differentiation capacity in retinoblastomas.^{21,35} His detailed work on immunohistochemical characteristics of different cell types of retinoblastoma (reactive astrocytes, undifferentiated neoplastic cells, differentiated cells forming Flexner-Wintersteiner rosettes) supported the view that retinoblastomas were composed of neuron-committed cells and favored the origin of these tumors from photoreceptor progenitor cells.⁵² Rubinstein correlated the neuropathological findings with the clinical picture and suggested a good prognosis for retinoblastomas; since then it has become known that retinoblastomas have a good response to treatment. Nevertheless, Rubinstein pointed out a very important aspect that clinicians should keep in mind; patients with retinoblastomas were prone to develop second malignancies, especially osteosarcoma, due to the genetic nature of heritable diseases.

Neurofibromatosis. Rubinstein defined the neuropathological features in 22 autopsies performed in cases of central and peripheral type neurofibromatosis. Besides the well-known cranial and spinal meningeal, nerve-sheath, and glial neoplasms in the central type of neurofibromatosis, he also defined very frequent and distinctive malformative CNS lesions (intramedullary and perivascular schwannosis, meningioangiomas, discrete ependymal ectopias, atypical glial cell nests in the gray matter, syringomyelia) associated with this type of the disease. In peripheral neurofibromatosis, he showed subependymal gliofibrillary nodules, hyperplastic meningioencephalic gliosis, and micronodular capillary and arteriolar proliferations typical of the vascular form of this disease.^{71,73}

Papillary Meningioma and Primary Meningeal Mesenchymal Chondrosarcoma. Rubinstein had a keen interest in meningiomas in addition to primary intraparenchymal brain tumors. He described papillary meningiomas and reported the largest series of these tumors, which are invariably associated with other histological features of malignancy. In his series, a relatively large proportion of papillary meningiomas occurred in children. The histological features were composed of a perivascular pseudopapillary pattern and local invasion with a high rate of recurrence and metastasis. The tumors often displayed aggressive clinical behavior marked by a high rate of local recurrence or the development of distant metastases.⁴³ Currently, in the World Health Organization classification,

papillary meningiomas are classified as Grade III, malignant meningiomas.

Rubinstein personally reviewed eight cases of primary meningeal mesenchymal chondrosarcoma and four similar cases previously reported by others. He defined the clinicopathological features. There was an apparent correlation between the frequency of mitotic figures and the likelihood of recurrence and metastasis. After his electron microscopic assessment he concluded that the neoplastic cells represented primitive precartilaginous mesenchyme displaying focal cartilaginous differentiation.⁸⁷

Sarcoma and Glioma. Rubinstein tried to explain the concurrent presence of gliomas and sarcomas in the CNS. He reported a series of mixed cerebral tumors histologically characterized by a peripheral distribution of the gliomatous elements in relation to a more centrally situated meningeal or intracerebral sarcoma. He suggested that these tumors be termed “sarcogliomas” to distinguish them from gliosarcoma.⁴¹ He also described mixed capillary hemangioblastoma and glioma and reserved the term “angioglioma” for only true mixed tumors of glial and vascular tissue origin.³

Hemangioblastoma. Rubinstein suggested that hemangiopericytoma, hemangioblastoma, or angioblastic meningioma were from the same source; from polyblastic mesenchymal cells originating in or derived from the meninges.²⁹ He maintained in culture a capillary hemangioblastoma resected from the vermis of a patient with von Hippel–Lindau disease and identified three cell types; endothelial cells, pericytes, and stromal cells, with their fine structural features and their architectural relationships to vascular lumina and to the extracellular space.⁸⁸ His observations of GFAP-positive astrocytes or astrocytic cell processes penetrating the margins of all the neuraxial tumors and none of the lesions occurring on nerve roots or the tumor explants maintained in an organ culture system, and also the presence of GFAP-positive stromal cells in hemangioblastomas, led him to develop the hypothesis that stromal cells were capable of taking up extracellular GFAP derived from the adjacent reactive astrocytes. He was the first to propose the concept of the uptake of GFAP by nonglial cells in the presence of dense gliosis.¹⁶

Malignant Peripheral Nerve Sheath Tumors. To understand the differentiation potential and biological behavior of malignant peripheral nerve sheath tumors, Rubinstein grew human nerve sheath tumors in organ culture systems; their sequential morphological features in vitro were compared with those of a human acoustic schwannoma maintained in similar culture systems. In organ culture systems, in which viable cultures were maintained up to 82 days, many of the experimental tumor explants exhibited progressive differentiation, with nuclear palisading, increasing whorl formation, and abundant reticulin fibers, and their pattern of histological organization came to resemble more closely that of the cultured acoustic schwannoma. The cultured experimental tumors infiltrated the sponge foam matrices, a feature that mimicked the invasive character of the original tumors in vivo.⁷⁸ Rubinstein studied the sequential electron microscopic features of both tumor cultures and showed that the malignant tumor cells had progressive elongation of their processes, with the development of an interdigitating pattern resembling that seen in well-differentiated schwannomas. The malignant tumor cells also

showed numerous micropinocytotic vesicles and various junctional complexes, which were characteristic of perineurial cells. He concluded that Schwann cells and perineurial fibroblasts were functional variants of the same cell type.¹² One pigmented malignant nerve sheath tumor was analyzed using light and electron microscopy, revealing melanosomes and premelanosomes in schwannoma-like cells, and it was concluded that neoplastic nerve sheath cells were capable of melanogenesis.⁸⁹

Radiation Effects on the CNS

Rubinstein considered many aspects of therapeutic ionizing radiation on the healthy brain or spinal cord. He described microscopic features: coagulative necrosis, demyelination, mononuclear response and perivascular lymphocytic infiltrate, fibrinoid necrosis of blood vessel wall, proliferation of endothelium, thrombotic occlusion, hyalinization of vessel wall, and extensive and relatively acellular fibrosis.⁷⁶ Rubinstein emphasized that those changes occurred in white matter; furthermore, he noted cortical changes including neuronal loss, degeneration, calcification, and bizarre and binucleated neurons. He recognized markedly thickened gyri with laminar disorganization and many unusually large and abnormally shaped ganglion cells. Abnormally large, misshapen neurons contained excessive accumulations of cytoskeletal intermediate filaments related to therapeutic irradiation of the brain.^{8,82}

Rubinstein was the first to describe radiation-induced changes in the cerebellum; the earliest change was the development of empty spaces in the Purkinje layer, confluence of those spaces to form vacuoles, loss of neurons, fibrillary gliosis, demyelination, extravasated fibrin material that spread radially along the molecular layer, and hyalinization of vessels. Rubinstein also reported secondary glial neoplasms, meningiomas, and neurofibrosarcomas following ionizing radiation therapy.²⁰

CONCLUSIONS

Lucien Rubinstein's remarkable career spanned four decades. Although he is most remembered for the classification of CNS tumors and publication of the authoritative textbook *Pathology of Tumours of the Nervous System*, he made invaluable contributions to modern neuro-oncology, including recognition of the importance of cell cultures and animal modeling, theories of tumorigenesis, description of new pathological entities, and careful classification of those previously known, by using innovative immunohistochemical techniques. Many of his correlations between the histopathological evaluations of tumors and outcome are just as valid today as when first proposed. His careful, systematic study of each tumor specimen and its relationship to the underlying neurobiological features set the standard for diagnosis and classification of CNS tumors.

References

1. Barnard R, Bigner DD: In memoriam Lucien J. Rubinstein, M.D. (1924–1990). *J Neurooncol* 9:III–VI, 1990
2. Bennett JP Jr, Rubinstein LJ: The biological behavior of primary cerebral neuroblastoma: a reappraisal of the clinical course in a series of 70 cases. *Ann Neurol* 16:21–27, 1984

3. Bonnin JM, Pena CE, Rubinstein LJ. Mixed capillary hemangioblastoma and glioma. A redefinition of the "angioglioma". **J Neuropathol Exp Neurol** 42:504–516, 1983
4. Bonnin JM, Rubinstein LJ: Astroblastomas: a pathological study of 23 tumors, with a postoperative follow-up in 13 patients. **Neurosurgery** 25:6–13, 1989
5. Bonnin JM, Rubinstein LJ: Immunohistochemistry of central nervous system tumors. Its contributions to neurosurgical diagnosis. **J Neurosurg** 60:1121–1133, 1984
6. Bonnin JM, Rubinstein LJ, Palmer NF, et al: The association of embryonal tumors originating in the kidney and in the brain. A report of seven cases. **Cancer** 54:2137–2146, 1984
7. Bonnin JM, Rubinstein LJ, Papasozomenos SC, et al: Subependymal giant cell astrocytoma. Significance and possible cytogenetic implications of an immunohistochemical study. **Acta Neuropathol** 62:185–193, 1984
8. Caccamo D, Herman MM, Urich H, et al: Focal neuronal gigantism and cerebral cortical thickening after therapeutic irradiation of the central nervous system. **Arch Pathol Lab Med** 113:880–885, 1989
9. Caccamo D, Katsetos CD, Herman MM, et al: Immunohistochemistry of a spontaneous murine ovarian teratoma with neuroepithelial differentiation. Neuron-associated beta-tubulin as a marker for primitive neuroepithelium. **Lab Invest** 60:390–398, 1989
10. Caccamo DV, Herman MM, Frankfurter A, et al: An immunohistochemical study of neuropeptides and neuronal cytoskeletal proteins in the neuroepithelial component of a spontaneous murine ovarian teratoma. Primitive neuroepithelium displays immunoreactivity for neuropeptides and neuron-associated beta-tubulin isotype. **Am J Pathol** 135:801–813, 1989
11. Caccamo DV, Herman MM, Rubinstein LJ: An immunohistochemical study of the primitive and maturing elements of human cerebral medulloepitheliomas. **Acta Neuropathol** 79:248–254, 1989
12. Conley FK, Rubinstein LJ, Spence AM: Studies on experimental malignant nerve sheath tumors maintained in tissue and organ culture systems. II. Electron microscopy observations. **Acta Neuropathol** 34:293–310, 1976
13. Curtis JL, Rubinstein LJ: Pigmented olfactory neuroblastoma: a new example of melanotic neuroepithelial neoplasm. **Cancer** 49:2136–2143, 1982
14. Davis RL, Kepes JJ, Rorke LB, et al: The art of brain tumor classification—a tribute to Lucien J. Rubinstein (1925–1990). Interview by Paul Kleihues. **Brain Pathol** 1:55–59, 1990
15. de Armond SJ, Eng LF, Rubinstein LJ: The application of glial fibrillary acidic (GFA) protein immunohistochemistry in neurooncology. A progress report. **Pathol Res Pract** 168:374–394, 1980
16. Deck JH, Rubinstein LJ: Glial fibrillary acidic protein in stromal cells of some capillary hemangioblastomas: significance and possible implications of an immunoperoxidase study. **Acta Neuropathol** 54:173–181, 1981
17. Eberhart CG, Burger PC: Anaplasia and grading in medulloblastomas. **Brain Pathol** 13:376–385, 2003
18. Eng LF, Rubinstein LJ: Contribution of immunohistochemistry to diagnostic problems of human cerebral tumors. **J Histochem Cytochem** 26:513–522, 1978
19. Erdelyi E, VandenBerg SR, Raese J, et al: Neural differentiation in the OTT-6050 mouse teratoma: enzymatic and immunofluorescence characterization of a tumor fraction showing melanogenesis in neuroepithelial cells. **Virchows Arch A Pathol Anat Histol** 393:27–37, 1981
20. Fontana M, Stanton C, Pompili A, et al: Late multifocal gliomas in adolescents previously treated for acute lymphoblastic leukemia. **Cancer** 60:1510–1518, 1987
21. Gass P, Frankfurter A, Katsetos CD, et al: Antigenic expression of neuron-associated class III beta-tubulin isotype (h beta 4) and microtubule-associated protein 2 (MAP2) by the human retinoblastoma cell line WERI-Rb1. A comparative immunoblot and immunocytochemical study. **Ophthalmic Res** 22:57–66, 1990
22. Goodrich LV, Milenkovic L, Higgins KM, et al: Altered neural cell fates and medulloblastoma in mouse patched mutants. **Science** 277:1109–1113, 1997
23. Halks-Miller M, Miller RG, Rubinstein LJ: Rat C-6 glioma cells maintained in an organ culture system: a study of kinetic parameters. **Cell Tissue Kinet** 14:59–72, 1981
24. Herman MM, Rubinstein LJ: Divergent glial and neuronal differentiation in a cerebellar medulloblastoma in an organ culture system: in vitro occurrence of synaptic ribbons. **Acta Neuropathol** 65:10–24, 1984
25. Herman MM, Sipe JC, Rubinstein LJ, et al: An experimental mouse testicular teratoma as a model for neuroepithelial neoplasia and differentiation. II. Electron microscopy. **Am J Pathol** 81:421–444, 1975
26. Herrick MK, Rubinstein LJ: The cytological differentiating potential of pineal parenchymal neoplasms (true pinealomas). A clinicopathological study of 28 tumors. **Brain** 102:289–320, 1979
27. Hess JR, Michaud J, Sobel RA, et al: The kinetics of human glioblastomas maintained in an organ culture system. An in vitro autoradiographic study. **Acta Neuropathol** 61:1–9, 1983
28. Horten BC, Rubinstein LJ: Primary cerebral neuroblastoma. A clinicopathological study of 35 cases. **Brain** 99:735–756, 1976
29. Horten BC, Urich H, Rubinstein LJ, et al: The angioblastic meningioma: a reappraisal of the nosological problem. Light-, electron-microscopic, tissue, and organ culture observations. **J Neurol Sci** 31:387–410, 1977
30. Ibayashi N, Herman MM, Boyd JC, et al: Kinetics and glial fibrillary acidic (GFA) protein production in a transplantable human giant cell glioblastoma (D-212 MG) of near haploid karyotype maintained in an organ culture system. An immunohistochemistry study. **Neuropathol Appl Neurobiol** 16:27–37, 1990
31. Ibayashi N, Herman MM, Boyd JC, et al: Relationship of invasiveness to proliferating activity and to cytoskeletal protein production in human neuroepithelial tumors maintained in an organ culture system: use of human cortex and dura as supporting matrices. **Neurosurgery** 26:629–637, 1990
32. Ibayashi N, Herman MM, Boyd JC, et al: Relationship of the demonstration of intermediate filament protein to kinetics of three human neuroepithelial tumor cell lines. Lack of neural-related proteins in most cells in S phase: a double-labeled immunohistochemical study on matrix cultures. **Lab Invest** 61:310–318, 1989
33. Kadin ME, Rubinstein LJ, Nelson JS: Neonatal cerebellar medulloblastoma originating from the fetal external granular layer. **J Neuropathol Exp Neurol** 29:583–600, 1970
34. Katsetos CD, Herman MM, Frankfurter A, et al: Cerebellar desmoplastic medulloblastomas. A further immunohistochemical characterization of the reticulin-free pale islands. **Arch Pathol Lab Med** 113:1019–1029, 1989
35. Katsetos CD, Herman MM, Frankfurter A, et al: Neuron-associated class III beta-tubulin isotype, microtubule-associated protein 2, and synaptophysin in human retinoblastomas in situ. Further immunohistochemical observations on the Flexner-Wintersteiner rosettes. **Lab Invest** 64:45–54, 1991
36. Kepes JJ: Pleomorphic xanthoastrocytoma: the birth of a diagnosis and a concept. **Brain Pathol** 3:269–274, 1993
37. Kepes JJ, Rubinstein LJ, Ansbacher L, et al: Histopathological features of recurrent pleomorphic xanthoastrocytomas: further corroboration of the glial nature of this neoplasm. A study of 3 cases. **Acta Neuropathol** 78:585–593, 1989
38. Kepes JJ, Rubinstein LJ, Eng LF: Pleomorphic xanthoastrocytoma: a distinctive meningocerebral glioma of young subjects with relatively favorable prognosis. A study of 12 cases. **Cancer** 44:1839–1852, 1979

39. Kleihues P, Cavenee WK: **Pathology and Genetics of Tumours of the Nervous System**. Lyon: IARC Press, 2000
40. Klein P, Rubinstein LJ: Benign symptomatic glial cysts of the pineal gland: a report of seven cases and review of the literature. **J Neurol Neurosurg Psychiatry** 52:991–995, 1989
41. Lalitha VS, Rubinstein LJ: Reactive glioma in intracranial sarcoma: a form of mixed sarcoma and glioma (“sarcoglioma”): report of eight cases. **Cancer** 43:246–257, 1979
42. Liwnicz BH, Rubinstein LJ: The pathways of extraneural spread in metastasizing gliomas: a report of three cases and critical review of the literature. **Hum Pathol** 10:453–467, 1979
43. Ludwin SK, Rubinstein LJ, Russell DS: Papillary meningioma: a malignant variant of meningioma. **Cancer** 36:1363–1373, 1975
44. Marsa GW, Goffinet DR, Rubinstein LJ, et al: Megavoltage irradiation in the treatment of gliomas of the brain and spinal cord. **Cancer** 36:1681–1689, 1975
45. Mork SJ, Rubinstein LJ: Ependymoblastoma. A reappraisal of a rare embryonal tumor. **Cancer** 55:1536–1542, 1985
46. Mork SJ, Rubinstein LJ: Metastatic carcinoma to glioma: a report of three cases with a critical review of the literature. **J Neurol Neurosurg Psychiatry** 51:256–259, 1988
47. Mork SJ, Rubinstein LJ, Kepes JJ: Patterns of epithelial metaplasia in malignant gliomas. I. Papillary formations mimicking medulloepithelioma. **J Neuropathol Exp Neurol** 47:93–100, 1988
48. Mork SJ, Rubinstein LJ, Kepes JJ, et al: Patterns of epithelial metaplasia in malignant gliomas. II. Squamous differentiation of epithelial-like formations in gliosarcomas and glioblastomas. **J Neuropathol Exp Neurol** 47:101–118, 1988
49. Nakagawa Y, Herman MM, Boyd JC, et al: Kinetics of the human medulloblastoma cell line, D283 Med, maintained in an organ culture system. Autoradiographic study after cumulative labeling in vitro: similarities to the kinetics of human medulloblastoma in situ. **Neuropathol Appl Neurobiol** 13:439–450, 1987
50. Nakagawa Y, Perentes E, Ross GW, et al: Immunohistochemical differences between intracranial germinomas and their gonadal equivalents. An immunoperoxidase study of germ cell tumors with epithelial membrane antigen, cytokeratin, and vimentin. **J Pathol** 156:67–72, 1988
51. Nakagawa Y, Perentes E, Rubinstein LJ: Immunohistochemical characterization of oligodendrogliomas: an analysis of multiple markers. **Acta Neuropathol** 72:15–22, 1986
52. Perentes E, Herbolt CP, Rubinstein LJ, et al: Immunohistochemical characterization of human retinoblastomas in situ with multiple markers. **Am J Ophthalmol** 103:647–658, 1987
53. Perentes E, Nakagawa Y, Ross GW, et al: Expression of epithelial membrane antigen in perineurial cells and their derivatives. An immunohistochemical study with multiple markers. **Acta Neuropathol** 75:160–165, 1987
54. Perentes E, Rubinstein LJ: Immunohistochemical recognition of human nerve sheath tumors by anti-Leu 7 (HNK-1) monoclonal antibody. **Acta Neuropathol** 68:319–324, 1985
55. Perentes E, Rubinstein LJ: Immunohistochemical recognition of human neuroepithelial tumors by anti-Leu 7 (HNK-1) monoclonal antibody. **Acta Neuropathol** 69:227–233, 1986
56. Perentes E, Rubinstein LJ: Recent applications of immunoperoxidase histochemistry in human neuro-oncology. An update. **Arch Pathol Lab Med** 111:796–812, 1987
57. Perentes E, Rubinstein LJ, Herman MM, et al: S-antigen immunoreactivity in human pineal glands and pineal parenchymal tumors. A monoclonal antibody study. **Acta Neuropathol** 71:224–227, 1986
58. Ramsay PB, VandenBerg SR, Eng LF, et al: Immunologic recognition of cell surface antigens in normal mouse neural tissues and in neuroepithelial cells of the OTT-6050 mouse teratoma. A radiometric, gel electrophoretic and morphologic (immunofluorescence and immunoperoxidase) study. **Acta Neuropathol** 56:214–224, 1982
59. Rawlinson DG, Herman MM, Rubinstein LJ: The fine structure of a myxopapillary ependymoma of the filum terminale. **Acta Neuropathol** 25:1–13, 1973
60. Rawlinson DG, Rubinstein LJ, Herman MM: In vitro characteristics of a myxopapillary ependymoma of the filum terminale maintained in tissue and organ culture systems. Light and electron microscopic observations. **Acta Neuropathol** 27:185–200, 1974
61. Ross GW, Rubinstein LJ: Lack of histopathological correlation of malignant ependymomas with postoperative survival. **J Neurosurg** 70:31–36, 1989
62. Rubinstein LJ: Correlation of animal brain tumor models with human neuro-oncology. **Natl Cancer Inst Monogr** 46:43–49, 1977
63. Rubinstein LJ: The correlation of neoplastic vulnerability with central neuroepithelial cytogeny and glioma differentiation. **J Neurooncol** 5:11–27, 1987
64. Rubinstein LJ: Cytogenesis and differentiation of pineal neoplasms. **Hum Pathol** 12:441–448, 1981
65. Rubinstein LJ: The definition of the ependymoblastoma. **Arch Pathol** 90:35–45, 1970
66. Rubinstein LJ: The development of contiguous sarcomatous and gliomatous tissue in intracranial tumors. **J Pathol Bacteriol** 71:441–459, 1956
67. Rubinstein LJ: Diagnostic markers in human neurooncology. A progress report. **Ann NY Acad Sci** 540:78–90, 1988
68. Rubinstein LJ: Discussion on polar spongioblastomas. **Acta Neurochir** 11 (Suppl 10):126, 1964
69. Rubinstein LJ: Embryonal central neuroepithelial tumors and their differentiating potential. A cytogenetic view of a complex neuro-oncological problem. **J Neurosurg** 62:795–805, 1985
70. Rubinstein LJ: Extracranial metastases in cerebellar medulloblastoma. **J Pathol Bacteriol** 78:187–195, 1959
71. Rubinstein LJ: The malformative central nervous system lesions in the central and peripheral forms of neurofibromatosis. A neuropathological study of 22 cases. **Ann NY Acad Sci** 486:14–29, 1986
72. Rubinstein LJ: Morphological problems of brain tumors with mixed cell population. **Acta Neurochir** 11 (Suppl 10):141, 1964
73. Rubinstein LJ: Pathological features of optic nerve and chiasmatic gliomas. **Neurofibromatosis** 1:152–158, 1988
74. Rubinstein LJ: Presidential address. Cytogenesis and differentiation of primitive central neuroepithelial tumors. **J Neuropathol Exp Neurol** 31:7–26, 1972
75. Rubinstein LJ: ‘Primitive neuroectodermal tumors’. **Arch Pathol Lab Med** 111:310–312, 1987
76. Rubinstein LJ: **Tumors of Central Nervous System**. Washington, DC: Armed Forces Institute of Pathology, 1972
77. Rubinstein LJ, Brucher JM: Focal ependymal differentiation in choroid plexus papillomas. An immunoperoxidase study. **Acta Neuropathol** 53:29–33, 1981
78. Rubinstein LJ, Conley FK, Herman MM: Studies on experimental malignant nerve sheath tumors maintained in tissue and organ culture systems. I. Light microscopy observations. **Acta Neuropathol** 34:277–291, 1976
79. Rubinstein LJ, Herman MM: The astroblastoma and its possible cytogenic relationship to the tancyte. An electron microscopic, immunohistochemical, tissue- and organ-culture study. **Acta Neuropathol** 78:472–483, 1989
80. Rubinstein LJ, Herman MM: A light- and electron-microscopic study of a temporal-lobe ganglioglioma. **J Neurol Sci** 16:27–48, 1972
81. Rubinstein LJ, Herman MM, Hanbery JW: The relationship between differentiating medulloblastoma and dedifferentiating diffuse cerebellar astrocytoma. Light, electron microscopic, tissue, and organ culture observations. **Cancer** 33:675–690, 1974
82. Rubinstein LJ, Herman MM, Miquel J, et al: The short- and long-term effects of ultraviolet irradiation on the exposed cat cerebrum. Light-microscopic, enzyme-histochemical and fine-structural observations. **J Neurol Sci** 13:351–375, 1971

83. Rubinstein LJ, Herman MM, VanderBerg SR: Differentiation and anaplasia in central neuroepithelial tumors. **Prog Exp Tumor Res** 27:32–48, 1984
84. Rubinstein LJ, Northfield DW: The medulloblastoma and the so-called “arachnoidal cerebellar sarcoma”. **Brain** 87:379–412, 1964
85. Russell DS, Rubenstein LJ: **Pathology of Tumours of the Nervous System**. London: Edward Arnold, 1959
86. Scheithauer BW, Rubinstein LJ: Cerebral medulloepithelioma. Report of a case with multiple divergent neuroepithelial differentiation. **Childs Brain** 5:62–71, 1979
87. Scheithauer BW, Rubinstein LJ: Meningeal mesenchymal chondrosarcoma: report of 8 cases with review of the literature. **Cancer** 42:2744–2752, 1978
88. Spence AM, Rubinstein LJ: Cerebellar capillary hemangioblastoma: its histogenesis studied by organ culture and electron microscopy. **Cancer** 35:326–341, 1975
89. Spence AM, Rubinstein LJ, Conley FK, et al: Studies on experimental malignant nerve sheath tumors maintained in tissue and organ culture systems. III. Melanin pigment and melanogenesis in experimental neurogenic tumors: a reappraisal of the histogenesis of pigmented nerve sheath tumors. **Acta Neuropathol** 35:27–45, 1976
90. Stanton C, Perentes E, Collins VP, et al: GFA protein reactivity in nerve sheath tumors: a polyvalent and monoclonal antibody study. **J Neuropathol Exp Neurol** 46:634–643, 1987
91. Urich H: Obituary. Lucien J. Rubinstein. **Neuropathol Appl Neurobiol** 16:371–372, 1990
92. Vandenberg SR, Herman MM, Rubinstein LJ: Embryonal central neuroepithelial tumors: current concepts and future challenges. **Cancer Metastasis Rev** 5:343–365, 1987
93. Vandenberg SR, Hess JR, Herman MM, et al: Neural differentiation in the OTT-6050 mouse teratoma. Production of a tumor fraction showing melanogenesis in neuroepithelial cells after centrifugal elutriation. **Virchows Arch A Pathol Anat Histol** 392:295–308, 1981
94. Vandenberg SR, May EE, Rubinstein LJ, et al: Desmoplastic supratentorial neuroepithelial tumors of infancy with divergent differentiation potential (“desmoplastic infantile gangliogliomas”). Report on 11 cases of a distinctive embryonal tumor with favorable prognosis. **J Neurosurg** 66:58–71, 1987
95. Vinore SA, Bonnin JM, Rubinstein LJ, et al: Immunohistochemical demonstration of neuron-specific enolase in neoplasms of the CNS and other tissues. **Arch Pathol Lab Med** 108:536–540, 1984
96. Vinore SA, Herman MM, Rubinstein LJ: Electron-immunocytochemical localization of neuron-specific enolase in cytoplasm and on membranes of primary and metastatic cerebral tumors and on glial filaments of glioma cells. **Histopathology** 10:891–908, 1986
97. Vinore SA, Herman MM, Rubinstein LJ: Localization of neuron-specific (gamma gamma) enolase in proliferating (supportive and neoplastic) Schwann cells. An immunohisto- and electron-immunocyto-chemical study of ganglioneuroblastoma and schwannomas. **Histochem J** 19:438–448, 1987
98. Vinore SA, Herman MM, Rubinstein LJ, et al: Electron microscopic localization of neuron-specific enolase in rat and mouse brain. **J Histochem Cytochem** 32:1295–1302, 1984
99. Vinore SA, Marangos PJ, Bonnin JM, et al: Immunoradiometric and immunohistochemical demonstration of neuron-specific enolase in experimental rat gliomas. **Cancer Res** 44:2595–2599, 1984
100. Vinore SA, Rubinstein LJ: Simultaneous expression of glial fibrillary acidic (GFA) protein and neuron-specific enolase (NSE) by the same reactive or neoplastic astrocytes. **Neuropathol Appl Neurobiol** 11:349–359, 1985
101. Wang VY, Zoghbi HY: Genetic regulation of cerebellar development. **Nat Rev Neurosci** 2:484–491, 2001
102. Wechsler-Reya RJ, Scott MP: Control of neuronal precursor proliferation in the cerebellum by Sonic Hedgehog. **Neuron** 22:103–114, 1999

Manuscript received February 25, 2005.

Accepted in final form March 30, 2005.

Address reprint requests to: Melike Mut, M.D., Department of Neurological Surgery, University of Virginia Health System P.O. Box 800212, Charlottesville, Virginia 22908. email: mm2ee@hsc mail.mcc.virginia.edu.