Workshop on cancer of the brain

Special review

DARELL D. BIGNER, M.D., PH.D., AND CHARLES B. WILSON, M.D.

Departments of Pathology (Neuropathology) and Experimental Surgery, and Virology, Duke University Medical Center, Durham, North Carolina, and Department of Neurological Surgery and Brain Tumor Research Center, University of California School of Medicine, San Francisco, California

The authors provide a summary of a workshop on “Cancer of the Brain.” This conference reviewed the current knowledge about the etiology and pathogenesis of human brain tumors and the experimental induction of comparable animal brain tumors, and considered new lines of research.

KEY WORDS • human brain tumor • carcinogen • tumor growth kinetics

A workshop on “Cancer of the Brain,” sponsored by the International Union Against Cancer, was held in Geneva, Switzerland, March 21-25, 1977. The purpose of the conference was to review the current knowledge about the etiology and pathogenesis of human brain tumors and the experimental induction of comparable animal brain tumors; to assess the information and techniques that are currently available from related fields and that could be applied to studies of human brain tumors; and to suggest and critique promising new avenues of investigation and research.

Fifteen investigators from a variety of disciplines participated in the workshop, including six neurobiologists, three neuropathologists, three cell biologists, a neuroembryologist, an epidemiologist, and a clinical neurosurgeon. The conference organizers, Professor Manfred Rajewsky, Dr. Ole D. Laerum, and Dr. J. R. DeLafresnaye of the International Union Against Cancer, had selected these participants on the basis of their knowledge and experience in their respective fields. Each participant presented to the meeting a 5- to 10-page manuscript, which was discussed in depth and criticized by the other participants. The final versions of these manuscripts were then edited by a subcommittee, who incorporated into them the consensus thinking and criticisms of the group. The final, compositely edited version of the conference proceedings will be published and distributed by the International Union Against Cancer in a single volume entitled “Cancer of the Brain,” scheduled to appear in the fall of 1977.*

Tumor Classification and Epidemiology

Professor K. J. Zülch (Cologne)

Professor Zülch made the opening address. His comments on the morphological

*The proceedings of the workshop, entitled “Cancer of the Brain,” can be obtained from Dr. J. F. DeLafresnaye, International Union Against Cancer, rue du Conseil-Général, 3, 1205 Geneva, Switzerland.
and functional classification of human brain tumors reflected his lifelong study and interest in the field. He presented a historical account of the classification of brain tumors, together with a courageous description of his own efforts over the past 10 years to gain an international consensus for adopting a classification scheme that would be uniformly acceptable to, and used by, pathologists throughout the world. With obvious joy and great pride, he presented and distributed to the group what is hoped will be a final consensus document and classification of brain tumors, which has been agreed upon by an international panel of neuropathologists under the auspices of the World Health Organization. It appears that all of the semantic arguments have been resolved through a series of practical compromises, and that the divergent views of Zülch, Rubinstein, and the Spanish School finally have been reconciled in a classification scheme that is acceptable to physicians of both the East and West.

Today, in the new era of clinical diagnosis and treatment of brain tumors that has arisen (largely through the strides that have been made in developing new diagnostic techniques, such as computerized tomography, and new therapeutic modalities, such as chemotherapy) this type of uniformly acceptable classification system is of paramount importance. An accurate classification, particularly of the malignant neuroectodermal brain tumors, greatly enhances the ability to predict survival; a precise classification of brain tumors is essential for a meaningful evaluation of new diagnostic and treatment modalities.

The uniform agreement among the neuropathologists present at the Geneva meeting, that detailed examination of large amounts of resected tumor is perhaps the most important prerequisite for both determining an accurate classification and assigning a postoperative prognosis, should be of particular interest to neurosurgeons.

Dr. L. C. Strong (Houston)

Dr. Strong described the epidemiology and genetic features of two neural tumors, retinoblastoma and neuroblastoma. Although these tumors rarely, and almost never primarily, involve the brain, Dr. Strong used them as examples to illustrate that distinct genes may predispose the organism to the development of a neural tumor. She cited evidence in support of the theory that a two-step mutation may be involved in the genesis of a tumor; in the case of hereditary tumors, genetic predisposition would constitute the first step, and a subsequent somatic mutation that triggers neoplastic transformation would constitute the second step. Although epidemiological information on brain tumors is scanty and largely anecdotal, retinoblastoma and neuroblastoma appear to have no counterpart in the brain. Still, the possibility that the development of some childhood tumors, such as medulloblastomas, may involve a genetic predisposition deserves consideration.

Viral and Chemical Carcinogens

There were three papers that discussed the possible viral or chemical carcinogenic etiology of human brain tumors and the induction of experimental brain tumors in animals by means of viral and chemical carcinogens.

Dr. D. D. Bigner (Durham)

Dr. Bigner reviewed the known oncogenic viruses that are capable of inducing experimental brain tumors. He stressed the susceptible species, the types of tumors induced, and virus-cell interrelationships in experimental brain tumors. Substantial progress has been made in this area within the past 10 years. The neuroblastic nature of rodent brain tumors induced with human adenovirus Type 12 has become recognized. Meningeal tumors have been induced with bovine papilloma virus. It has been found that simian vacuolating virus 40 (SV-40) induces choroid plexus papillomas rather than ependymomas, as had formerly been thought. Expanding numbers of tumors, including medulloblastomas and pinealomas, have been induced with the human papova virus isolates (MAD-1, 2, 3, 4, and 5) from progressive multifocal leukoencephalopathy. Bona fide anaplastic astrocytomas have been induced with avian sarcoma virus (ASV), and quantitative principles of virology have been applied to induce anaplastic astrocytomas with ASV in large numbers of animals. Studies have achieved such uniform mortality distributions (survival curves) with these
Workshop on cancer of the brain

animals that chemo- and immunotherapy now can be evaluated, relative to control animals, in a primary tumor system before clinical application. Anaplastic astrocytomas and glioblastomas have been induced using murine sarcoma virus and simian sarcoma virus. Finally, experimental trials have not yet shown that brain tumors can be induced with herpes viruses.

Despite the wealth of data on experimental brain tumors induced with viruses, there is no convincing evidence to date of any virus involvement in the etiology of human brain tumors. The most promising studies, which must be confirmed in future work, are the detection of SV-40 T antigen in meningiomas, and the continued search for additional associated cases of astrocytomas in cases of progressive multifocal leukoencephalopathy.

Professor P. Kleihues (Freiburg)

Professor Kleihues reviewed the induction of experimental brain tumors with the topical application of chemical carcinogens, but his talk focused on the voluminous work with the 30 compounds that, over the past 10 years, have been shown to induce a high incidence of nervous-system tumors in animals following systemic application. The most effective of these compounds are the N-nitrosamides; in particular, the nitrosourea derivatives, the dialkyl-aryl triazenes, and the azo, azoxy, and hydrazo compounds. He reviewed the metabolism of these compounds, highlighting the specific, known mechanisms by which they ultimately become carcinogens through the action of electrophilic reactants formed during their metabolism in vivo.

He also presented an extensive review of his own and others' work on DNA modification and repair by alkylating carcinogens, which indicated that several hypotheses about the mechanism of action of nitrosoureas in preferential tumor induction in the nervous system now can be refuted. He discussed the work showing that there is no preferential accumulation of the carcinogen in the nervous system relative to other organs, as well as some of the cytostatic lethal effects of the loss of alkylated DNA bases. The repair of specific sites of DNA damage, such as repair excision of O\(^{-}\)-alkyl guanine from DNA, has been shown to correlate with the target organ site of carcinogenesis. For example, rats have high repair rates for O\(^{-}\)-methyl guanine in the liver, but when the brain is the oncogenic target organ the rate of repair is much lower. Nevertheless, the repair of O\(^{-}\)-methyl guanine proceeds at the same rate of the brain for two mouse strains that differ significantly in their susceptibility to nervous system tumor-induction with methylnitrosourea (MNU).

Thus, Kleihues concluded that, despite significant progress in understanding some factors about the molecular mechanism of DNA damage and repair of chemical carcinogens that are selectively neurotropic, much remains to be explained. There is no uniform concept that explains the high susceptibility of the developing nervous system to this class of neuro-oncogenic agents, and no explanation for the fact that the perinatally administered ethylating agents are considerably more effective than the methylating carcinogens. Moreover, although a steadily growing list of chemical compounds have been positively identified as causative agents in systemic human cancer, the pathogenesis and etiology of brain tumors in humans remains obscure.

Dr. O. D. Laerum (Bergen) and Professor M. F. Rajewsky (Essen)

Dr. Laerum and Professor Rajewsky focused on the interval between initial carcinogen-cell interaction and the first detection of neoplastic growth. They pointed out that chemical carcinogens, in addition to their oncogenic action, exert dose-dependent cytotoxic and cytocidal effects on target cell populations. The cytocidal effects may be important in carcinogenesis, since a rebound increase in the rate of DNA replication and cell division often follows cytotoxicity resulting from nitrosourea and other neuro-oncogenic compounds. Widespread degenerative changes in perivascular cells, glial cells, and oligodendrocytes follow systemic administration of nitrosoureas.

It is difficult to study the interval following cell-carcinogen interaction in neoplastic transformation in vivo, since the earliest unequivocal neoplastic changes following nitrosamide administration appear in 20 days, and changes often do not appear for up to 140 days. Laerum and Rajewsky have developed a combined in vivo-in vitro method to study neural cells during this "latent period" that precedes bona fide neoplastic
Pregnant rats were given oncogenic doses of ethylnitrosourea (ENU) at 18 days of gestation; 20 to 90 hours after a transplacental pulse of 75 μg of ENU/gm, the brains were placed into culture. A characteristic sequence of phenotypic alterations was observed. During early primary growth, a mixture of glial-like cells and epithelioid cells was present. Glial-like cells appeared between the 10th and 40th day of in vitro growth, whereas these cells were not observed in control cultures. From the 40th to 100th day, fully proliferating, glial-like cells aggregated into foci. In the fourth stage, between 100 and 200 days after culture was initiated, morphological transformation occurred; there was loss of anchorage dependency, the cells could form colonies in semi-solid agar, and they became tumorigenic in rats. Although a characteristic set of phenotypic changes existed long before tumorigenic cells were present, bona fide tumorigenic cells were not observed until after 100 to 200 days in culture. In comparison with in vivo methods alone, the in vivo-in vitro method offers the distinct advantage of permitting continual observation; nonetheless, much knowledge remains to be gained about the initial events that occur between carcinogen exposure and neoplastic transformation.

**Tumor Growth Kinetics**

Professor B. Schultze-Maurer (Würzburg)

Professor B. Schultze-Maurer described the kinetic characteristics and proliferative potential of cells in the brains of fetal and postnatal mammals. The data were obtained in mice and rats using ³H-labeled thymidine and high resolution autoradiography. The various cell types were divided into two groups on the basis of their proliferative properties. In the first group (neurons, microglia, pericytes, ependymal cells, and epithelial cells of the choroid plexus), proliferation occurs primarily in the prenatal period, but may continue at a greatly reduced rate during the first 2 to 3 postnatal weeks. In adult animals, neurons, pericytes, and microglia are incapable of proliferation, whereas ependyma, and possibly choroidal epithelium, show very slight proliferative activity and maintain the potential for renewed proliferation under special circumstances. Cells of the second group (astroglial, oligodendrogial, and endothelial cells) continue to proliferate at a slow rate throughout adult life; since all three types have remarkably similar and short cell cycles (cell cycle time of 20 hours), this process evidently serves to balance a naturally occurring low rate of cell loss, and maintain a constant cell population.

Of particular interest were two areas in which proliferation continues in the postnatal period: the external granular layer of the cerebellum, where neuronal proliferation continues until the third postnatal week in the rat; and the subependymal layer of the lateral ventricles, where neuroglial proliferation continues throughout adult life. Cells migrate out of the subependymal layer, presumably during the process of normal replacement of astrocytes and oligodendrocytes.

Professor R. Balázs (Carshalton)

Professor Balázs reviewed the current knowledge, much of it originating in his laboratory, concerning the effects of hormones and neurohumoral agents on the developing rat brain. Neonatal thyroid deficiency reversibly retarded cell acquisition in the cerebellum; however, the forebrain was not affected. In the cerebellum, a prolonged period of cell proliferation in the external granular layer ultimately compensated for the reduced rate of cell acquisition. Administration of thyroid hormone caused premature cessation of cell proliferation throughout the brain, and resulted in selective acceleration in the rate of cell acquisition in the cerebellum during the first postnatal week.

The administration of adrenal corticosteroids severely reduced the normal increase in the number of brain cells throughout the period of treatment and, in contrast to the reversible effect with thyroid deficiency, a permanent deficit occurred because of inadequate compensation. Similarly, because germinal cells in the brain are as vulnerable as other cells in the body, undernutrition during early development caused an irreversible depression in cell acquisition. Balázs introduced the possibility that neurotransmitters can act as neurohumors to control cell replication in the brain. He cited experiments in which reserpine had profound inhibitory effects on the developing brain, such as lengthening the cell cycle time, and he encouraged further investigation of drugs that affect neurotransmitters as modulators of cell proliferation.
Workshop on cancer of the brain

proliferation in the developing and adult brain. Because the factors that lead to accelerated cell proliferation also may be involved in the evolution of tumors, the proposed studies have distinct oncogenic implications.

Professor C. G. Steel (London)

Professor Steel reviewed growth-kinetics studies on the more common non-neural tumors, and examined the relationship of the brain-tumor data to this much larger body of information. He questioned the DNA synthesis time (Ts) of 9 hours for glioblastomas reported by Hoshino, et al., because longer S-phase durations have been obtained in other human tumors when more accurate techniques, such as labeled mitoses, have been used. Assuming that Ts is 15 hours for glioblastomas, Steel estimated a potential doubling time for 10 days. Because this value is inconsistent with clinical observations, he also assumed a high rate of cell death and cell loss, in addition to the low-growth fraction (proportion of cycling cells in the tumor cell population) reported by Hoshino, et al.钢 Steel considered the cell-survival studies of Rosenblum, et al., to be consistent with clonogenic survival data on a variety of other experimental tumors.

In no other variety of cancer is there such a clear difference between the kinetics of tumor tissues and the coexisting normal tissues. Steel concluded that, although this difference in cell kinetics favors a chemotherapeutic approach to brain tumors, the pharmacokinetic problems of drug access create an overriding limitation on the success of chemotherapy at the present time.

Animal Models for Tumor Research

Dr. C. B. Wilson (San Francisco)

Dr. Wilson gave an overview of the animal models that are available for laboratory research. Although no one of these numerous models perfectly represents spontaneous human tumors, different models have specific advantages for particular types of studies. Despite an expanding array of autochthonous and transplantable tumors induced by chemical carcinogens and oncogenic viruses, three currently available models are particularly adaptable to therapeutic experiments: 1) the polycyclic hydrocarbon-induced, transplantable, murine ependymoblastoma and derived cell lines; 2) the nitrosourea-induced transplantable rat tumors; and 3) the avian sarcoma virus (ASV)-induced autochthonous rat tumors. The ependymoblastoma models are used for drug screening studies requiring large numbers of animals. The MNU-induced rat gliosarcoma model is advantageous because it is adaptable to a refined clonogenic cell assay for quantitative cell kill experiments and studies of post-treatment kinetics. The ASV model developed by Bigner and Swenberg most nearly resembles human glioblastoma, and has the added advantage that its anatomical characteristics simulate spontaneous tumors, as opposed to implanted tumors.

He concluded that the ideal tumor system would be both autochthonous and transplantable, as well as astrocytic and capable of parallel in vitro cultivation. While the assumption that such a system would predict clinical responsiveness is open to question, the experimental advantages of a tumor that mimics human anaplastic astrocytomas is obvious. Although gaining clearer insight into the predictive value of current models should be given priority over developing new models for therapeutic experiments, newer models must be developed; particularly necessary are well differentiated, “pure” neuroglial tumors of different cell types, a transplantable medulloblastoma, and reproducible models in larger animals, including subhuman primates. He suggested that an understanding of the reasons for our clinical failures is no less important than an understanding of our successes, and that significant progress in the treatment of human tumors seems unlikely until more successful strategies are developed, based upon the information that can be obtained in animal model systems.

Cell Interactions

Two of the neurobiologists focused their presentations on cell-cell differentiation and interaction in the brain.

Dr. N. W. Seeds (Denver)

Dr. Seeds described some of the cell interactions, beginning with those that take place during early neuroembryogenesis. In the early developing embryo, the neural plate
is induced by underlying chordomesoderm, which presumably is chemical in nature; the embryonic ectoderm produces only epithelium and no neural plate. It is presumably under a similar, specific chemical control, the nature of which is presently unknown, that clonal specification takes place, with subsequent proliferation and migration of specific neuronal and glial cell populations. Finally, following cell migration and cessation of mitotic division of cells in established anatomical locations, axonal growth and final positioning of perikaryon takes place.

Growth cones of neural fibers appear to possess specific likes and dislikes, in that they will attach and grow over certain cells, but rapidly retract and change direction upon contact with others. Synapse formation occurs when appropriate recognition events take place. The roles of other specialized cell-cell interactions, such as tight junctions and desmosomes, have not been explored sufficiently. Synaptogenesis is a complex process that involves cell migration to the correct topographical position, morphological extension of processes to appropriate interaction points, and biochemical differentiation with synthesis of specific enzymes and cell surface receptors necessary for functional activity. The signals controlling these processes are not well understood, but it may be that sprouting and re-enervation of damaged axonal fibers are influenced by molecules such as nerve growth factor. Myelination, which may involve both secretory components and cell surface recognition molecules, is another process that should lend itself to this line of investigation.

There are two highly successful experimental approaches to the study of cell-cell interactions. One is the in vivo use of neurologically mutant mice that have malpositioned cells for comparison with normal animals. Another is the use of reaggregation cell cultures that mimic morphogenesis, an area in which Seeds and Moscona have been leaders. With cultures such as these, work now has begun to show that reaggregation of cells from specific brain regions is temperature-dependent, which implies that recognition is an active mechanism, and not a simple static lock-key relationship. Moreover, experiments with specific chemical agents that prevent aggregate formation, such as colchicine, and agents that interfere with the sorting out process in aggregates, such as bromodeoxyuridine, indicate that the use of reduced variables that can be controlled in culture systems should make a systematic analysis of specific molecules on cell-cell interactions possible in the future.

The investigation of specific surface molecules that may influence the recognition processes between nervous system cells because of chemical, topographical, and temporal differences in their distribution on the cell surface is also now approachable by a direct analysis of membrane proteins, and by immunological analysis for cell surface differences. A better understanding of cell-cell interaction would have direct application to neoplasia, because knowledge about the signal for cessation of cell division in germinal zones of the nervous system could provide a basis for both the understanding and control of the neoplastic cell. The uncontrolled growth of neoplastic cells and their invasive characteristic might be related to changes in the specific cell recognition molecules in the nervous system.

**Professor D. Monard (Basel)**

Professor Monard reviewed the literature and reported on his own work with glial cell influences on neuronal cells. He emphasized that, except for the well established function of glia in myelin formation, there is a tremendous lack of knowledge regarding glial-neuronal cell function. He also emphasized that the modulation of neuronal activity by glia probably requires an uptake of ions and neurotransmitters, a release of neurohumoral factors, degradation of toxic substances, or a membranal interaction, any of which probably are localized to specific sites in the brain, and that, consequently, very low concentrations of the molecules would be involved. His own experimental approach has been to study the effect of factors elaborated by glia that produce changes in neurons or glial cells in cell culture. He reviewed cellular interactions involving low-molecular substances, and used as examples the accumulation of cyclic AMP in C-6 glioma cells following an increase in norepinephrine, isoproterenol, and epinephrine levels in extracellular culture fluid.

Another category that Monard discussed was the substances with low molecular weight that affect such membranal interactions as changes in the expression of nervous system-
specific cell-surface antigens in the presence of one molar dibutyryl cyclic AMP. As an example of non-neuronal regulation of neurotransmitter synthesis, he presented data to show that the synthesis of acetylcholine from radioactive choline by neurons is increased up to 1000-fold when the neurons are cocultured with non-neuronal cells.

Finally, he described the glial-neuronal interactions that affect neuronal membrane plasticity. Monard has detected a macromolecular factor released by glial cells in culture which induced morphological differentiation of neuroblastoma cells and which, he believes, is immunochemically and biochemically distinct from nerve growth factor. He concluded that cell culture systems afford an excellent opportunity to obtain molecular information about glial-neuronal interactions.

**Phenotypic Markers of CNS Cells**

Phenotypic markers of normal and neoplastic nervous system cells were discussed by the other three neurobiologists.

**Dr. H. R. Herschman (Los Angeles)**

Dr. Herschman described the origin of a number of ENU-derived tumors of the central nervous system in the rat, the phenotypic assessment of the "neuronal" or glial nature of those cell lines, and his view of their potential utility in neurobiology and oncology. In addition, he described alternatives to the two standard laboratory cell lines, namely, the murine C-1300 neuroblastoma as a neuronal representative, and subclones of the rat C-6 glioma cell line as a glial representative, that have been used for most of the studies in neurobiology over the last decade.

Five research groups, those of Schubert, Fields, Laerum and Rajewsky, West and Herschman, and Pfeiffer and Wechsler, have developed large numbers of tumors and clones from nervous system tumors induced transplacentally with ENU in rats. Of the 22 cell lines developed by Schubert, five had electrically excitable membranes; none of the lines developed by Laerum and Rajewsky were excitable. Schubert considered his lines to be "neuronal." A re-examination of Schubert's electrophysiologically positive cell lines with a biochemical ion-flux assay correlated well with electrophysiological data. Eight of the 35 cell lines Herschman tested by the ion-flux methods were also positive; on the basis of the sodium ion-flux data, those eight lines have been tentatively considered to be neuronal.

Both Schubert's and Herschman's cell lines have been examined quantitatively for S-100 and 14-3-2 brain-specific proteins. Both laboratories concluded that these antigens are valuable for distinguishing neural from nonneuronal cells, but are of little use in distinguishing "glial" from "neuronal" cells. Herschman examined his cell lines for cortisol-inducible glycerol phosphate dehydrogenase and found that eight lines were positive, but the ion-flux assays showed that among those eight were three that had evidence of neuronal nature. These dual biochemical criteria made Herschman suspect that some of his cells had both neuronal and glial properties, a possibility that is consistent with transformation of "stem" cells during transplacental ENU carcinogenesis.

There is great controversy among the laboratories of Schubert, Herschman, and Laerum and Rajewsky about the interpretation of low levels of neurotransmitter synthesis in the rat nervous-system cell lines. In contrast to the low activities found in rat lines, the appropriate neurotransmitter enzymes in adrenergic and cholinergic murine and human neuroblastomas have high specific activities. Schubert's cell lines also apparently have cell surface antigens that allow glial cells to be distinguished from neuronal cells.

Five human neuroblastoma lines have been studied by Herschman using the ion-flux assay. Four of the five lines were positive; two were "adrenergic," that is, had high levels of tyrosine hydroxylase; a third line produced very high levels of acetylcholine. These lines are now being utilized to evaluate the use of nerve growth factor, 6-hydroxydopamine, neurotransmitters, and other agents as potential therapeutic agents for human neuroblastoma. A careful biochemical characterization of the phenotypic properties of nervous-system tumor cells may be a prelude to the development of rational, biochemical approaches to treatment.

**Professor B. Hamprecht (Munich)**

Professor Hamprecht described some of the biochemical markers in normal and neoplastic cell populations of the mammalian...
nervous system, in clonal cell lines derived from nervous system tumors, in primary cultures of dissociated nervous tissue, and in reaggregation cultures of dissociated nervous tissue. In order of specificity as to “glial cell” identification, Hamprecht felt that two of the most specific biochemical markers were glycerol phosphate dehydrogenase induction by hydrocortisone and glial fibrillary acidic protein. Other, but not necessarily specific, markers expressed in glial cells are S-100 protein, 2’3’-cyclic AMP, 3’-phosphohydrolase, carbonic anhydrase, and glycogen content. Myelin basic protein-like bands on polyacrylamide gels have been found in membrane extracts of a few experimental glioma cell lines, but there have been no reports of an independent identification of myelin basic protein by more definitive biochemical or immunochemical properties. Other substances that have yet to be proven of value as glial markers are uptake of neurotransmitters and receptors for neurohormones.

One of the more reliable identifications for neuronal cells in culture is believed to be the synthesis of neurotransmitters, especially such enzymes as choline, acetyl transferase, and one or several of the enzymes of the noradrenaline synthetic pathway, such as tyrosine hydroxylase, dopa-decarboxylase, and dopamine-β-hydroxylase. Other identifying aspects of neuronal cells are the electrical and chemical excitability of the membranes, the capacity to form synapses, and the presence of receptors for neurohormones. Both Hamprecht and Herschman summarized their findings by suggesting that, while a number of markers, such as the inducibility of glycerol 3-phosphate dehydrogenase, the presence of glial fibrillary acidic protein or S-100 protein, or the presence of enzymes specific for synthesis of neurotransmitters, such as choline acetyltransferase or tyrosine hydroxylase, are good means for differentiating neural from non-neural tissue, the capability for distinguishing neuronal and glial populations and sub-populations from one another on a biochemical basis is, at present, far from ideal.

Drs. C. Goridis and M. Schachner (Boston)

Drs. Goridis and Schachner discussed recent progress in immunological markers of normal and neoplastic brain cells. Immunological methods already have begun to define cell surface antigens and molecules that may be important in cell-cell interaction in the nervous system. These findings not only may be valuable for neurophysiological and neuroembryological applications, but also, more practically, for clinical applications in the immunodiagnosis and possible immunotherapy of brain tumors.

A number of laboratories have taken a serological approach to distinguishing nervous system surface antigens. Basically, this approach involves immunization with whole cells, crude particulate matter, or clonal cell lines in syngeneic or xenogeneic animals, absorption of the non-nervous-system specificities with non-neural tissues to remove non-specific antibodies, and demonstration of the nervous-system specificity of the remaining antibody population by testing a broad panel of nervous-system and non-nervous-system cells. The assay systems that have commonly been used are complement-dependent cytotoxicity assays and membrane immunofluorescent assays. A few workers have used sensitive radioimmunoassays, as well. Among the specific antigens that have been detected, perhaps the most significant is the Thy-1 antigen, which is shared by T-thymocytes and mouse brain. The exact cellular and subcellular locus of Thy-1 is not known at the present time. Normal mouse H-2 and human HLA histocompatibility antigens are present in brain.

Goridis and Schachner also presented a detailed discussion of the surface antigens that have been defined on mouse brain and some mouse brain tumors. NS-3 is present on C-1300 mouse neuroblastoma cells, normal mouse brain, and kidney. NS-1 and NS-2 are brain-specific and present on murine ependymoblastomas, some other murine glioblastoma cell lines, and on brain. Other surface antigens that are shared by brain and embryonic tissues, such as those designated NS-4 and NS-5, are present on the cerebellum, retina, sperm, and kidney, but not on any other adult tissues, were also described.

They concluded that “although few of the presently available antisera show the desired specificity needed for classifying and manipulating distinct cell populations, the rapid progress made over the last few years has justified the hope that an array of antigenic surface components will be defined, which may eventually serve as specific
Workshop on cancer of the brain

markers not only for given types, but also for certain differentiation events." If these hopes are realized, then practical clinical immunodiagnostic and immunotherapeutic methods might also be developed.

Comments

This workshop brought up a number of points of which clinical neurosurgeons should be aware. Perhaps most importantly, there are exciting studies being made in the basic areas of neuroscience and neurobiology, many of which could now be evaluated in pilot clinical studies in patients with brain tumors.

Several areas of progress and insight were defined by the conference. The new and, it is hoped, internationally acceptable uniform classification scheme for brain tumors is a significant accomplishment. The knowledge about molecular mechanisms of carcinogenesis with systemically administered chemicals is increasing, and may shed as much light on mechanisms of action of brain tumor chemotherapeutic agents as on mechanisms of carcinogenesis. Since etiological (viral and chemical) studies still have offered no clues that promise the prevention of brain tumors in the near future, there is a continuing need for better methods for treatment of malignant brain tumors. The increasing understanding of cell-cycle events in normal and neoplastic glial and neuronal cell populations should be helpful in tailoring more effective use of currently available chemotherapeutic agents. At least three good animal models of human brain tumors for therapeutic experimentation now are available. Neoplastic glia potentially can be controlled by applying variations of the natural regulatory signals of cell division, differentiation, migration, and recognition. And finally, the possibilities for diagnosing tumors and following tumor size have been expanded by the clinical investigation of serum or cerebrospinal fluid levels of biochemical markers of normal and neoplastic glia, such as glial fibrillary acidic protein, S-100 protein or immunologically distinctive glial cell surface antigens.

It has become apparent that there is far too little communication between basic neurobiologists, cell biologists, oncologists, and those in the clinical neurosciences who each day are diagnosing and treating patients with brain tumors. This type of workshop should be only the first of many such endeavors to rectify this situation. Those of us who wish to see this fruitful interaction between workers in the clinical and basic neurosciences develop further must redouble our efforts toward achieving a vital, active communication.

References


Address reprint requests to: Darell D. Bigner, M.D., Department of Pathology, Box 3156, Duke University Medical Center, Durham, North Carolina 27710.