Capillary Physiology and Drug Delivery in Central Nervous System Lymphomas

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To evaluate whether the chemosensitivity of primary central nervous system lymphomas to water-soluble drugs could result from improved drug delivery, we quantitatively assessed pharmacokinetic factors in seven patients. The capillary permeability surface product was found to be significantly increased in central nervous system lymphomas compared with glioblastoma multiforme, medulloblastomas, and metastases. Tumoral blood flow was significantly greater than in normal white matter. Our results suggest favorable pharmacokinetics to water- and lipid-soluble drugs in primary central nervous system lymphomas.

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Primary central nervous system (CNS) lymphomas represent a unique entity with a biological behavior conspicuously different from other primary brain tumors, especially malignant gliomas, and also from secondary brain tumors. In contrast with other adult brain tumors and in agreement with systemic lymphomas, primary CNS lymphomas are chemosensitive to water-soluble agents, namely methotrexate.[1,2] Using high-dose methotrexate (up to 8gm/m^2), we found that approximately 70% of patients respond. The neuroradiological appearance of primary CNS lymphomas is different from other brain tumors, with the majority of cases showing homogeneous contrast enhancement.[3,4]

Both the imaging appearance and the responsiveness to methotrexate indicate favorable pharmacokinetics for water-soluble drugs. A quantitative assessment of vascular physiological parameters such as capillary permeability, regional blood volume, and regional intratumoral blood flow would allow analysis of the pharmacokinetics of methotrexate in individual tumors and may also lead to the development of treatments aiming at the vasculature of primary CNS lymphomas.

This is still needed because even the best currently available treatments are not curative, with median survival plateauing at approximately 40 months. We therefore conducted a study of the capillary physiology of primary CNS lymphomas using quantitative measurements of physiological parameters in untreated, biopsy-proven primary CNS lymphomas.

Patients and Methods

Seven patients harboring primary CNS lymphoma verified by serial stereotactic biopsies were enrolled in this study. The study was approved by the local ethics committee, and informed consent was given by all patients. All patients had not received steroids for a minimum of 7 days at the time of investigation, which was performed under stereotactic conditions. Four patients (Patients 1, 3, 6, and 7) had never been exposed to steroids. There were two female and five male patients (age: range, 16–69 years; median, 48.3 ± 15.4). All patients had B-cell lymphomas, as proved by immunohistochemistry using antibodies against common B-cell antigens (L26). Patient characteristics are detailed in Table 1.

Measurements of Capillary Permeability and Vascular Volume

Using contrast-enhanced dynamic computer tomography (Siemens Somatom HiQ; Siemens Medical Systems, South Iselin, NJ), we measured the blood-to-tissue transfer constant, K_1, the tissue-to-blood efflux rate, k_2, and the vascular plasma volume, V_P, using iopamidol as marker substance (molecular weight = 777; octanol/water partition coefficient = 0.0038). Because this substance does not enter cells and is not metabolized during the 25-minute measurement period,[5] the amount of contrast medium, $A_m(t)$, at each image location can be described by a two-compartment model resulting in:

$$A_m(t) = V_p C_a(t) + K_1 \int_0^t C_a(\tau) e^{k_2(\tau - t)} d\tau \qquad \text{(Eq.1)}$$

In this model, which has been validated against the "gold standard" quantitative autoradiography in experimental autochthonous gliomas using alpha-amino-isobutyric acid (AIB)[5] and human brain tumors, the blood-to-tissue transfer constant (K_1) equals the capillary permeability surface product as a first approximation.[6]

A 5-minute infusion of 2ml iopamidol (330mg/ml) per kilogram body weight was performed during which 12 computed tomography (CT) scans at an identical position through the maximum diameter of the tumor were performed (25-second interscan delay). Scanning was performed after cessation of the infusion for another 20 minutes with a 99-second interscan delay. Scans were performed with a slice thickness of 5mm at 133keV, 475mAs, and a 2.2-millisecond pulse width using a 512 × 512 matrix. The

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Table 1. Patient Characteristics (tumor area reflects size of tumor as seen in axial slice through maximum diameter)

Patient	Sex	Age (yr)	Tumor Location	Tumour Area (cm^2)	Histology
1	F	63	Right frontal	53.9	B cell
2	F	67	Right frontal	31.6	B cell
3	M	48	Left occipital	10.1	B cell
4	F	38	Splenium	60.0	B cell
5	M	21	Left occipital	29.4	B cell
6	M	28	Left frontal	40.6	B cell
7	M	54	Left parietal	6.3	B cell

arterial input curve, $C_a(t)$, was measured from a major vessel from CT and normalized against the plasma concentration determined by blood samples taken before and 10 minutes after infusion. The parameters K_1, k_2, and V_p then were calculated at each image location by a least squares fit of the equation to the time course of CT numbers.

Regional analysis was performed from the parameter images after superimposing the stereotactic trajectory to relate histological findings from the serial biopsy sites with imaging findings. Regions of interest for whole tumor were chosen by visual outlining of the contrast-enhancing mass. The regions of interest then were checked against the histological findings from those areas to ensure that vital tumor was present in these regions. Region of interest analysis was performed separately by two investigators (P.C.W., K.K.) with an interobserver variability of less than 3%.

Measurement of Blood Flow

Regional cerebral blood flow was measured using stable-Xenon CT (Messer/Griesheim) with a 6-minute wash-in protocol and a modified Kety–Schmidt equation.[7] A concentration of 30% xenon, 30% O_2, and 40% air was used for all studies. Again a Siemens Somatom HiQ (Siemens) was used for all blood flow studies. Scanning parameters were 80keV and 500mAs, with a slice thickness of 10mm. End-tidal O_2, CO_2, respiratory rate, and volume were continuously monitored. Using commercial software (Siemens), we generated parameter maps for blood flow from the CT images, and regional cerebral blood flow was determined for tumor and normal contralateral gray and white matter. Again, the whole tumor was outlined on the superimposed contrast image.

Results

In this group of untreated primary CNS lymphomas, capillary permeability surface product showed a widespread variation with values from 16.1 ± 14.2 to 43.5 ± 30.2μl/gm/min. As can be seen from the parameter image (Fig), there was also a significant intratumoral variability of capillary permeability. The distribution of permeability values within the tumor resembles an almost Gaussian distribution. k_2 values were found to vary from 0.011 to 0.133L/min^{-1}. The vascular volume showed variation between 0.003 and 0.065ml/gm. Table 2 shows the values for all seven patients. The mean value of 29.5 ± 10.6μl/gm/min for K_1 in this tumor group (whole-tumor values) reflects the increased capacity for passive diffusion of iopamidol in CNS lymphomas.

Blood flow ranged between 35.7 and 61.8ml/100gm/min, with a mean value of 43.16 ± 10.5ml/100gm/min. Normal gray and white matter values in these patients were 57.1 ± 5.2 and 20.7 ± 2.5ml/100gm/min, respectively; both were significantly different from the tumor value ($p < 0.05$; t test). No correlation existed between K_1 and V_p or tumor size ($p > 0.29$; Kendall's τ).

Discussion

This is the first series of measurements of capillary physiology in untreated primary CNS lymphomas using a CT-based method rendering a high spatial resolution. Previous measurements in partially treated patients using positron emission tomography have resulted in inconclusive findings with two patients characterized as having K_1 values of only 2 and 8μl/gm/min before treatment.[8] It also remains unclear whether the patients were taking steroids. In a comparative study with identical methodology, we found that anaplastic astrocytomas have a mean K_1 value of 12.3 ± 7.9μl/gm/min, medulloblastomas 10.5 ± 6.3μl/gm/min, and glioblastomas 11.2 ± 5.4μl/gm/min, and metastasis showed a mean K_1 value of 16.5± 11.8μl/gm/min. Thus, primary CNS lymphomas are significantly more permeable than other primary and secondary brain tumors ($p < 0.01$; t test), which might form the basis for their therapeutic responsiveness to high-dose methotrexate chemotherapy.[9] Although methotrexate has a lower molecular weight (MW = 454) and a lower octanol/water partition coefficient (log o/w = −2.52) than iopamidol, the differences are minor and have no major pharmacokinetic impact.[10] Therefore, iopamidol is well suited to mimic the transport of methotrexate across the blood–brain barrier (BBB).

The rather large variability in vascular volume seen in our patients with limited variability as to their capillary permeability can most likely be attributed to their variable degree of vascular endothelial growth factor expression, as shown in a correlative study of permeabil-

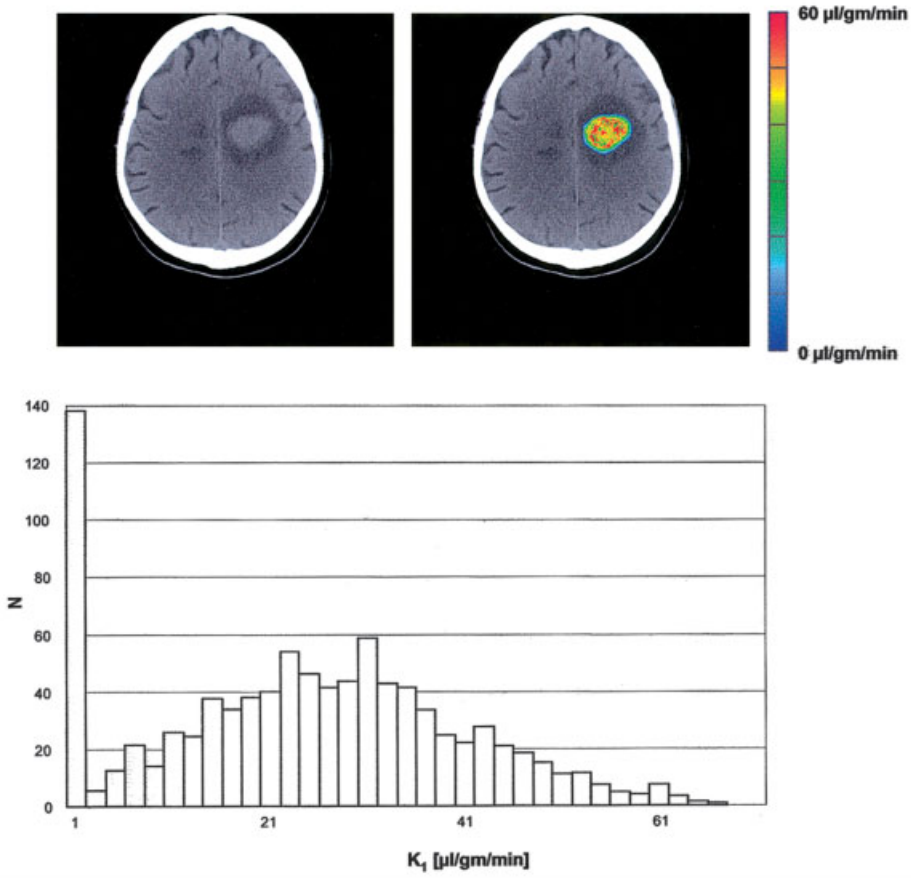

Fig. Color-coded representation and histogram of distribution of K_1 *values (blood-to-tissue transfer constant) obtained in a primary central nervous system lymphoma. The corresponding native computed tomography scan is shown.*

ity, vascular space, and vascular endothelial growth factor expression in brain tumors.[11]

Our measured values for K_I in CNS lymphomas actually approach those of extracerebral lymphomas measured with the same technique, which were in the range of 36.4 to 198.5µl/gm/min.[12] Methotrexate is an extremely water-soluble compound that barely crosses the BBB even at high doses and also poorly diffuses into other tumors with low capillary permeability. Although CSF levels of methotrexate can be obtained and have been measured with methotrexate, they do not reflect extracellular space concentrations. With primary CNS lymphomas being much more permeable to water-soluble compounds than other brain tumors, methotrexate can more easily diffuse into the extracellular space of primary CNS lymphomas where it is readily taken up by the cells. Our study also raises the question as to the general necessity of hyperosmotic BBB disruption in the treatment of primary CNS lymphomas. In the absence of Class I or II evidence for

Table 2. Physiological Parameters in Primary Central Nervous System Lymphomas (rCBF in Patient 6 could not be acquired for technical reasons)

Patient	K_1 (µl/gm/min)	Vp (ml/gm)	k_2 (l/min)	rCBF
1	31.4	0.007	0.029	35.7
2	37.5	0.004	0.032	47.4
3	16.3	0.002	0.011	32.9
4	36.2	0.065	0.086	61.8
5	16.1	0.03	0.062	37.7
6	43.5	0.033	0.133	
7	25.3	0.047	0.130	43.5
Mean	29.5 ± 10.6	0.027 ± 0.024	0.069 ± 0.049	43.16 ± 10.5

rCBF = regional cerebral blood flow.

BBB disruption, this study could lay the foundation for a controlled study including permeability measurements to clarify the value of BBB disruption in CNS lymphomas.[13]

Because blood flow also is quite high compared with other primary and secondary brain tumors, although not influencing the overall extraction of water-soluble compounds from the plasma space into the extracellular space, primary CNS lymphomas may also be ideal targets for treatment with lipophilic drugs such as temozolomide.[14]

Our data show that primary CNS lymphomas are a different physiological entity compared with other brain tumors and render a basis for the development of rational treatment schedules using the unique physiology of these tumors. It also allows quantifying treatment effects especially those of glucosteroids on these tumors in a reproducible fashion. In conjunction with measurements of methotrexate levels, deriving an area-under-the-curve extraction fraction calculation for methotrexate can be performed and intratumoral drug concentrations can be estimated.

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Pick Bodies in a Family with Presenilin-1 Alzheimer's Disease

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Presenilin-1 (PS-1) mutations can cause Pick's disease without evidence of Alzheimer's disease (AD). We describe a family with a PS-1 M146L mutation and both Pick bodies and AD. Sarkosyl-insoluble hyperphosphorylated tau showed three bands consistent with AD, although dephosphorylation showed primarily three-repeat isoforms. M146L mutant PS-1 may predispose to both Pick's disease and AD by affecting multiple intracellular pathways involving tau phosphorylation and amyloid metabolism.

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Presenilin-1 (PS-1) mutations account for most familial Alzheimer's disease (AD) and have been reported in familial frontotemporal dementia (FTD), although most reports lack pathological confirmation. Dermaut and

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