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Plasminogen activator and hemorrhage in brain tumors

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Introduction

The incidence of spontaneous intracranial hemorrhage caused by brain tumors has been documented to be 1.3% to 15% [1–6] and has been correlated with histological types. Wakai *et al.* [7] reported that the incidence of hemorrhage from various types of brain tumors was found to be 100% in both choriocarcinoma and embryonal carcinoma, 33% in adenoid cystic carcinoma, 25% in pituicytoma, 20% in mixed glioma, 16.7% in choroid plexus papilloma, 15.8% in pituitary adenoma, 8.8% in ependymoma, 7.8% in glioblastoma, 7.0% in oligodendroglioma, 4.5% in astrocytoma, 3.3% in craniopharyngioma, 2.9% in metastatic tumor, 1.6% in medulloblastoma, and 1.3% in meningioma. Other authors reported hemorrhage with metastatic melanoma [8] and neurinoma [9, 10]. The types of hemorrhage from brain tumors, except for pituitary adenoma, were intratumoral (66.7%), intracerebral (20%), subarachnoid (20%), and subdural (2%) [7].

Postoperative hemorrhage and brain tumors

At one time, neurosurgeons frequently encountered intracerebral hemorrhage in the operative field and in operative sites following surgery. However, with the advent of microsurgical procedures, the incidence of postoperative intracranial hemorrhages has dropped considerably from the incidence reported earlier. In 1985, Fukamachi *et al.* [11] reported that of 1074 operations, 116 (10.8%) showed hemorrhages on CT. Eighty-three of 349 patients with brain tumors had associated intracerebral hematomas, including 51 small, 25 medium, and 7 large hemorrhages. Thirty-two medium and large hematomas were ascribed to 12 meningiomas, 5 glioblastomas, 5 pituitary adenomas, 3 metastatic tumors, 2 low-grade gliomas, 2 acoustic neurinomas and 3 others. Subsequently, Kalfas and Little [12] reported their experience with postoperative intracranial hematoma during a period of 11 years. Of 4992 intracranial procedures, 40 patients (0.8%) experienced postoperative symptoms or neurological findings secondary to an intracranial hematoma. Hematomas in 33 patients occurred at the operative site. Brain tumor was the reason for operation in 23 (56%) of the patients who developed a clot. Meningioma was the most prevalent tumor (n = 9), for which 8 patients underwent gross total removals. Glioblastoma (n = 2), anaplastic glioma (n = 2), and pituitary adenoma (n = 2) were the only other tumor types that occurred more than once in this series. Coagulopathy (thrombo-cytopenia, anticoagulent therapy with warfarin) was present in three of the 40 patients who developed postoperative hematomas; the remaining 37 cases had normal coagulation parameters prior to the procedure. Postoperative intracerebral hematomas have been correlated with most histological types of brain tumors, especially meningioma and glioblastoma.

Causes of hemorrhage from brain tumors

Intracranial hemorrhage in patients with brain tumors suggests two separate etiologies. The first is a spontaneous intracranial hemorrhage from brain tumors. The patient's symptoms and signs correspond to the pathological entity (i.e., hypertensive intracerebral hematoma, subarachnoid hemorrhage, intratumoral hemorrhagic, or subdural hematoma). The second is a hemorrhage tendency. Coagulation disorders may occur during protracted intracranial operations, or an unexpected hemorrhage in operative fields may be encountered during and/or after surgery.

In general, hemorrhage from brain tumors has been believed to be caused by the rupture of thin-walled fistulous or friable large vessels, by infarction, or by the rupture of small arteries and arterioles invaded by tumor cells [13–15]. Involvement of the fibrinolytic system has rarely been considered as an initiating factor in hemorrhage from brain tumors. Recently, however, involvement of coagulation and fibrinolysis in patients with brain tumors is being related to the concept of tumor-host interaction, and its acknowledgment is being well-established.

Historical background of brain tumors and fibrinolysis

Early studies of brain tumors and thromboembolic and hemorrhagic tendencies began with an emphasis on the general factors involved in the tumor-host interaction. Initially, Morozov [16, 17] reported that patients with benign brain tumors showed a tendency toward a reduction in the coagulant properties of the blood without change in the fibrinolytic activity, whereas patients with malignant tumors had a moderate increase in the fibrinolytic activity of the blood. Burgman *et al.* [18] reported that patients with glial tumors had an increase in the blood coagulation properties. Moreover, Scharrer and Hubner [19] pointed out that the fibrinolytic activity was mildly elevated in patients with primary brain tumors and was moderately reduced in patients with metastatic tumors. From these results, it is difficult to determine the extent to which coagulation and fibrinolysis are involved in brain tumors.

We [20–22] took notice of fibrinolytic abnormalities due to an increase of plasma fibrinolytic activity appearing in three of 13 patients with meningiomas prior to, during, or after surgery. These patients demonstrated hemorrhagic diathesis at the incision site that was associated with an increase of fibrin/fibrinogen degradation products (FDP) concentration and a decrease of fibrinogen concentration in plasma. Antiplasmin drugs (gabexate and tranexamic acid) were effective in minimizing blood loss both during and after operation. These patients showed pre- and postoperative elevations of tissue-plasminogen activator (t-PA) related to fibrinolytic activity in euglobulin fractions (EFA). Fibrin autography revealed that a broad lytic band of mol wt 50–60 kDa, probably free t-PA, appeared in the plasma obtained from two of the three patients after operation when EFA was significantly elevated. In all patients, the t-PA antigen levels were normal preoperatively, but increased both during and after operation, and correlated mainly with intensities of a lytic band of mol wt 110 kDa, probably t-PA complex with plasminogen activator inhibitor-type 1 (PAI-1). Microscopic sections of the tumor showed meningotheliomatous meningiomas with an angiomatous component in three patients. These results suggest that in the patients with meningiomas, the excessive fibrinolysis induced the local hemorrhagic diathesis during operation and was related to plasma t-PA.

Sawaya *et al.* [23] measured a complete preoperative fibrinolytic profile, including total fibrinolytic activity, t-PA, plasmin inhibitor, PAI-1, protein C, and plasminogen, in 114 patients with various brain tumors. Plasminogen and plasmin inhibitor were decreased in 15% of the patients with malignant brain tumors. Tissue-PA was abnormally low in several patients and in almost 40% of the patients with brain metastasis. PAI-1 was above the upper limit of normal in approximately 50% of the patients, but particularly in patients harboring gliomas, glioblastomas, and metastases. They suggested that the significant changes in the fibrinolytic parameters could not be totally related to the histological nature of the tumor and should be correlated with

other biological parameters, including the tumor's content in fibrinolytic and coagulation promoting substances and the condition of the surrounding brain.

From a viewpoint of local fibrinolysis and tumor-host interface, Bock *et al.* [24] and Tovi *et al.* [25] studied local fibrinolysis using fibrin plate assay and Todd's method. The fibrinolytic activities of the dura mater, cerebral tissue, and various brain tumors were measured to determine the causes of delayed coagulation in the course of neurosurgery of long duration. A high content in PAs was noted in meningioma, medulloblastoma, cerebellar sarcoma, and glioblastoma. Koos *et al.* [26] noted that proteinase inhibitor (Trasylol) proved efficacious in prophylaxis and for treatment of pre- and postoperative hemorrhage that occurred in conjunction with neurosurgical procedures. This inhibitor was thought to inhibit free plasmin and other substances with thromboplastic action released in great amounts in the course of brain surgery, as well as the kinines that are discharged through the proteolytic activity of kininogens. Subsequently, Sawaya and Highsmith [27] studied a total of 58 fresh human brain tumor samples using a zymographic assay technique. They described higher PA activity and various molecular weight patterns in malignant tumors. It is generally thought that local fibrinolysis is increased in malignant brain tumors.

Fibrinolytic sequence and inhibitors in brain tumors

The serine protease plasmin is generated from plasminogen by the action of t-PA or uPA. Histochemical and immunocytochemical techniques have t-PA localized in endothelial cells of veins and some, but not all, smaller arteries. Soreq and Miskin [28] detected PA activity in the brains of mice and rats that was found to be associated with cell bodies in neuronally-enriched region; with endothelial, meningeal, and ependymal layers; and with granular neurons in the developing cerebellum. Zymography showed that the majority of PA activity in the brain was of the t-PA type. Danø *et al.* [29] also noted that uPA immunoreactivity was not detected in either the brain or the pituitary gland, but it was demonstrated by an immunofluorescence method with monoclonal antibodies [30] in a human glioblastoma cell line.

Tissue-PA is produced by vascular endothelial cells, whereas uPA is produced and secreted by cells in solid tumors as a single-chain enzymatically-inactive proenzyme form (pro-uPA) that may bind to specific receptors on the cell surface [29, 31, 32]. Pro-uPA is transformed into the enzymatically-active two-chain high molecular-weight form (HMW-uPA). HMW-uPA converts plasminogen to plasmin, which then degrades the fibrin-fibrinonectin portion of the tumor stroma [33] and activates procollagenase IV to enzymatically-active collagenase IV [34]. In that way, destruction of the surrounding brain tissue and the tumor tissue itself may be supported and may result in intratumoral and peritumoral hemorrhage or infarction.

Hemostasis in the surrounding brain

Hemostasis in the brain requires the activation of the tissue factor (TF, so-called tissue thromboplastin)-dependent pathway. The first step in the activation of factor VII to factor VIIa occurs during normal hemostasis only after factor VII binds to TF. The second step for the normal operation of the TF-dependent pathway is the activation by factor VIIa/TF complexes of two substrates, factor IX and factor X. The third step involves the direct activation of factor X by factor VII/TF. It also involves an indirect activation of factor X, an essential process for normal hemostasis in the brain, by factor IXa/factor/VIIIa/phospholipid complex. The indirect activation of factor X is essential.

The brain contains a large amount of TF, and human TF antigen expression by immunohistochemistry has been observed as a diffuse staining throughout the brain parenchyma [35, 36]. Recently, Eddleston *et al.* [37] reported that analysis of murine brain sections by *in situ* hybridization demonstrated high levels of TF mRNA in cells that expressed glial fibrillary acidic protein, a specific marker for astrocytes. Primary mouse astrocyte cultures and astrocyte cell lines from mouse, rat, and

human constitutively expressed TF mRNA and functional protein. They proposed that astrocytes forming the glia limitans around the neural vasculature and deep into the meninges were intimately involved in controlling hemorrhage in the brain.

Conclusion

In summary, it is believed that tumor hemorrhage is likely the result of local disturbances in hemostasis. Such disturbances are caused, at least in part, by the established imbalances in the fibrinolytic factors contained in the brain tumor tissue itself.

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