LECTURE

Intraoperative Navigation and Fluorescence Imagings in Malignant Glioma Surgery

Masahiro Toda

Department of Neurosurgery, Keio University School of Medicine, Tokyo, Japan

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Abstract: The current basic goals in glioma surgery are radical tumor resection without triggering the development of new neurological deficits. Although complete removal of malignant gliomas is extremely difficult because of the tumors' infiltrative characteristics, gross total resection of the tumor is known to be associated with improved outcome in the patients. To enable safe and radical resection of malignant gliomas, especially those adjacent to eloquent brain areas, rapid progress has been made in the development of operative support techniques; e.g., navigation systems to provide information about the anatomical and functional locations in the brain and a fluorescence imaging technique for differentiating brain tumors from normal brain tissue. These intraoperative imaging techniques in glioma surgery have improved the functional outcomes of glioma patients. (Keio J Med 57 (3) : 155–161, September 2008)

Key words: glioma, imaging, navigation, fluorescence, surgery

Introduction

Gliomas are the most common malignant brain tumors. With the currently available treatment modalities for malignant gliomas, the median survival still remains less than 2 years.[1] Gliomas are characterized by an infiltrative nature, and extensive invasion into the surrounding normal brain tissue is frequently seen. Even though the margin between the lesion and the normal brain is clearly delineated on magnetic resonance images (MRI), it is histologically obscure in most malignant glioma cases.

A basic goal of current glioma surgery is maximum tumor resection without triggering the development of new neurological symptoms. Although complete removal of malignant gliomas is extremely difficult, reduction of the tumor volume nonetheless decreases the compression of the surrounding normal brain tissue and increases the therapeutic efficacy of adjuvant therapies, including radiotherapy and chemotherapy. A correlation has actually been reported between the tumor resection rate and the prognosis in patients with malignant gliomas.[2,3]

Accurate intraoperative determination of the location of the tumor and of the eloquent areas in the brain may improve the rate of tumor resection. With the remarkable advances in diagnostic imaging techniques, including computed tomography (CT) and MRI, computerized navigation systems have been developed, that allow real-time information to be obtained about the positions in the operative field.[4–7] Research on the mapping of brain functions has also progressed, and it is now possible to obtain information about locations associated with important functions (movement, speech, etc.).[8–16] Although still in the clinical research stage, there have also been some attempts to differentiate infiltrative tumors from the normal brain tissue using fluorescent dyes and excitation lights.[17–22] This article provides an outline of the navigation systems that provide information about the anatomical and functional locations in the brain during surgery, and of a fluorescent imaging technique which biologically identifies malignant brain tumors, with reference to our experience.

Intraoperative Navigation in Image-guided Brain Tumor Surgery

In brain tumor surgery, accurate orientation is required

Corresponding Author: Masahiro Toda, M.D. Department of Neurosurgery, Keio University School of Medicine, 35 Shinanomachi, Shinjuku-ku, Tokyo 160-8582, Japan, Phone: 81-3-5363-3587, Fax: 81-3-5363-3862, E-mail:todam@sc.itc.keio.ac.jp

for tumor removal without interfering with important brain structures. The surgical navigation system is an imaging support system which provides information about positions in the actual operative field by coordinate representation, and the positions are confirmed intraoperatively by their reflection on the images.[4,5,7] With this system, data, including preoperative CT or MRI findings, are reconstructed on multiplanar 2-D images, i.e. axial, coronal and sagittal slices, and the sites indicated with a pointer probe are displayed in real-time on the images. Using this system, surgery can be performed while ascertaining the 3-D orientation of the operative field, which allows more accurate and safer surgical maneuvers.

Navigation systems

There are several types of navigation systems in terms of the detection method: e.g., the machine arm type, in which the distance between the probe tip and the starting point of the arm and the angle are calculated and displayed, the magnetic field type, in which a magnetic field develops in the operative field, based on which the position of the probe tip can be detected, and an optical type, which is equipped with an infrared camera. The optical type of navigation system is currently the most widely employed, because of its high accuracy and relatively easy operability. We are currently using the StealthStation (Medtronic Sofamor Danek, Co., Ltd.), which is an optical type of navigation system.[2,3]

The StealthStation consists of a Workstation with a monitor which displays the navigation images and an infrared camera which detects the position of the tip of a pointer probe (Figure 1). The equipment includes a reference frame as an optical reference point for navigation and a pointer probe for confirmation of the position. Both have built-in light-emitting diodes (LED), including the active type, with a cord, which emits light, and the passive type, without a cord, which is equipped with a reflective ball and utilizes reflected light from an infrared lamp source. The device also includes an adapter, the "Sure Trak System", which allows surgical equipment to be used instead of the pointer probe. The system allows recognition of positional relations of the built-in LED or the reflective ball attached to the tip of the equipment and implementation of navigation. The adapter system is especially useful for transnasal surgery in deep narrow operative fields and for endoscopic surgery through a one bar-hole in the ventricles in the brain.

The position is measured with an infrared camera according to the principle of triangular measurement, and information about the position of the pointer probe tip is displayed in real time on the images, obtained preoperatively (Figure 1). The advantage of the optical type of navigation system is that the accuracy of navigation is maintained, independently of the surgical position or movement of the infrared camera, because the reference frame is integrated with the patient.

Application of the navigation system to brain tumor surgery

Markers for registration are applied to scalp sites, and the necessary MRI and/or CT imaging are conducted before the operation. Routine axial images are obtained, and the image information is loaded onto the StealthStation in the DICOM III format. Basically, the reference frame is connected to the head frame for registration after the surgical position has been confirmed in the operating room. Another system for direct immobilization of the reference frame to the head is also being developed for cases in which head frame is not used, such as in the case of transnasal surgery. The data for registration are input by applying the pointer probe tip to the scalp. There are 2 methods for registration in the StealthStation: point registration, in which the actual head is adjusted to the image data set up by using the markers on the scalp before imaging, and surface registration, in which the data collected at 40 or more points on the scalp surface are randomly input to obtain the 3-D form. Stable registration with high accuracy can be maintained using these two registration methods.

When the pointer probe is applied to the operative field, the probe tip position is displayed in real time on the monitor, allowing confirmation of the orientation. The display monitor can indicate multiplanar images of axial, coronal and sagittal slices. Virtual extension of the tip on the software gives feedback on the position of any point on the prolonged line of the probe. Therefore, it is possible, as a matter of course, to let the virtual extension of the tip invade the skull even before craniotomy. This method provides for precise designing of the minimum skin incision and craniotomy even prior to the brain tumor surgery.

The navigation system is mainly used to confirm, intraoperatively, the location of tumors and of important functional areas, such as the motor and language areas, in the brain. Recently, identification of the eloquent areas has become possible with the development of non-invasive techniques to analyze brain functions, such as functional MRI and magnetoencephalography (MEG).[8,10,14] Utilization of these functional examinations allow important brain areas to be located even prior to the surgery. By means of the navigator, tumors and eloquent areas mapped on MRI images are accurately projected onto the brain surface during the operation.

Thus, more accurate surgery has become possible with the use of navigation systems. Actually, it has been reported that intraoperative neuronavigation in glioma surgery improves functional outcomes of glioma patients.[24,25] However, there are still problems that remain to be resolved. The biggest of these is that navigation

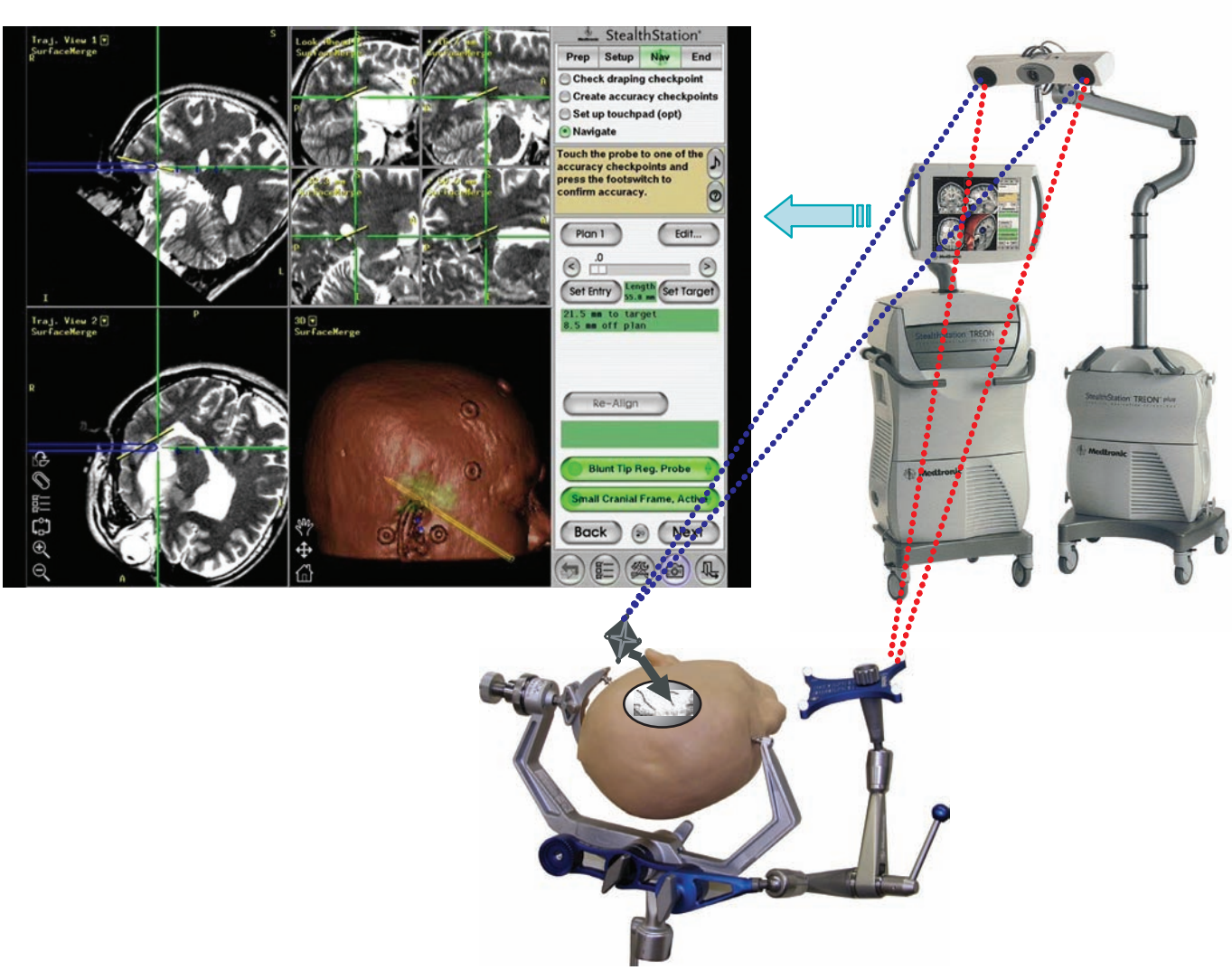

Fig. 1 Optical navigation system in brain tumor surgery. The infrared camera detects points of a pointer probe and a reference frame as an optical reference point for navigation. The position of the pointer probe tip is displayed in real-time on images obtained preoperatively.

surgery can only minimally deal with 'brain shift'. When the arachnoid membrane is incised during the operation, cerebrospinal fluid flows out, causing brain deformity. Some patients with severe brain edema may have marked brain shift at the time of incision of the dura mater. As a matter of course, compression of the brain during a surgical procedure also causes considerable displacement. Thus, navigation surgery using preoperative imaging findings as reference cannot deal adequately with such considerable brain shifts. To overcome this problem related to brain shift, we confirm the position of the tumors and important areas by navigation as early as possible after craniotomy, and insert tubes at the margin of the tumor using the navigation system (Figure 2). Then, tumor removal is performed using the anchored tubes as indicators.

As a countermeasure against brain shift, there have also been some attempts to incorporate intraoperative ultrasonography into navigation and to display preoperative images of the same cross-sectional surfaces. By means of a position detector attached to the ultrasonic probe, it may become possible to confirm the degree and range of tumor removal with an echographic system and to compare the findings with preoperative images. Re-examination by MRI intraoperatively may also allow the problem posed by such brain shifts to be overcome. Techniques for MRI in the operating room are being actively developed,[26–28] and real-time imaging of lesions, which allow changes along with the surgical maneuvers to be imaged, have become possible. Fusion with such intraoperative image updating techniques would allow more accurate navigation to be conducted.[29,30]

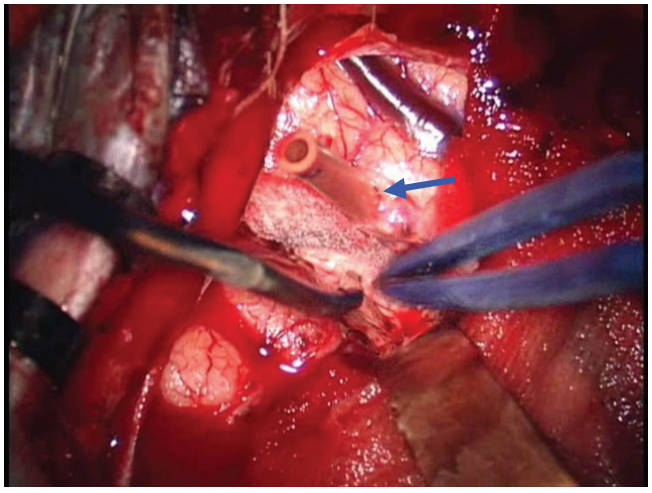

Fig. 2 Image-guided glioma surgery using landmark-tubes. As a countermeasure against brain shift during operation, landmark-tubes are inserted at the margin of tumor by navigation before tumor removal.

Intraoperative Fluorescence-guided Surgery of Brain Tumors

The boundary between malignant gliomas, which are infiltrative tumors, and the surrounding brain tissue is poorly defined, and it is difficult to determine tumor margins during operation. Until now, evaluation of tumor margins has been largely dependent on the surgeon's experience. In an attempt to delineate the margin between normal brain and tumor, a technique called intraoperative photodynamic diagnosis (PDD) has been developed.[17,20,21,31,32] In this method, a fluorescent substance is incorporated into brain tumor, and the fluorescence detected during the operation allows the normal brain tissue to be differentiated from the tumor tissue.

A basic principle underlying this method is that a fluorescent dye administered into the body leaks out of the tumor blood vessels due to the disrupted blood-brain barrier (BBB), which, when excited with an appropriate wavelength of light, emits fluorescence which can be observed in the operative field through a filter. However, this technique has a disadvantage that the dye may leak out and emit fluorescence in the absence of the BBB even at non-tumor sites. Recently, 5-aminolevulinic acid (5-ALA), a precursor of heme, has been applied for PDD in glioma patients;[18,20,21,32] 5-ALA, which induces the synthesis of protoporphyrin IX (Pp IX) in the mitochondria, has been shown to be incorporated into tumor cells when administered orally. When the substance is irradiated with a violet-blue excitation light, the tumor cells emit red fluorescence that can be observed through a filter.

Fluorescent dyes

1) Fluorescein sodium

Fluorescein sodium has been used in the field of ophthalmology, mainly for fundus fluorescein angiography. It also has a long history of use in the field of neurosurgery; even as early as in 1948, Moore *et al*. had applied it to glioma surgery.[33] Malignant gliomas show marked hypervascularity at the margin of the tumor, which has a high density of tumor cells. Since these blood vessels are deficient in normal BBB, fluorescein sodium (molecular weight, 332) easily passes through the blood vessels. The blood vessels at the center of the tumor also differ from normal blood vessels, and do not have a BBB. Under these circumstances, the use of fluorescein sodium is appropriate for PDD in glioma surgery; since fluorescence has been recognized to be consistent with the area of enhancement on CT or MRI images, removal of the part emitting fluorescence allows removal of the tumor itself.[34]

2) 5-Aminolevulinic acid (5-ALA)

5-ALA is a natural biochemical precursor of heme that induces the synthesis of the fluorescent molecule, Pp IX, through metabolic conversion in the mitochondria.[35] 5-ALA (molecular weight, 167.6) given via the oral route normally does not enter the normal brain tissue, because of the presence of the BBB. However, it can easily pass through the disrupted BBB in the glioma tissue.[21] Since Pp IX emits peak fluorescence at 635 nm with a peak excitation wavelength at 405 nm, it is observed as a red light through a filter that allows this wavelength to pass. Clinical use of 5-ALA for photodynamic therapy (PDT) or PDD has also been reported for lung cancer, ovarian cancer, and bladder cancer.[36–38] In the USA, it has been used with the approval of the Food and Drug Administration (FDA) as a therapeutic drug for solar keratosis, while its use has not yet been approved by the pharmaceutical authority in Japan, and it is only used after obtaining approval of the ethics committee of the respective institutes. The tumor tissue concentration of the fluorescent dye peaks at 2-6 hours after oral administration, and disappears by 12 hours. Administration of antacid should be avoided at the time of oral administration of the dye, because the dye is easily decomposed in the presence of alkali.

Use of 5-ALA is contraindicated in cases of porphyria, a genetic disease. Pp IX biosynthesized from 5-ALA is a phototoxic substance, although the incidence of skin photosensitivity is lower than that reported for conventional porphyrin derivatives.[20] Since the administered 5-ALA is excreted into the urine within 24 hours after oral administration and does not remain in the skin, the occurrence of photosensitivity can be ade-

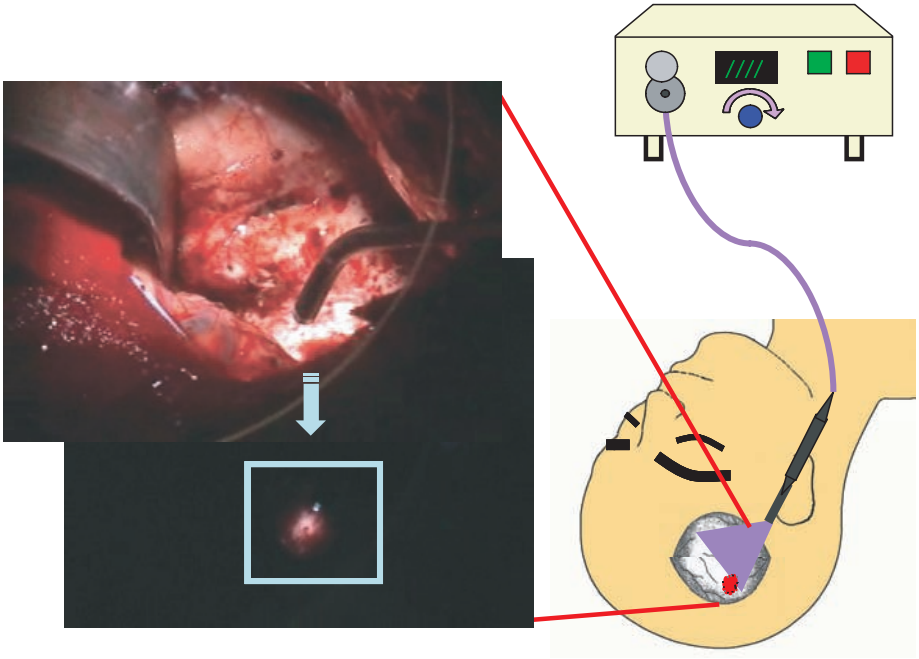

Fig. 3 Fluorescence-guided glioma surgery using 5-ALA
Red fluorescence emitted by protoporphyrin IX, that is biosynthesized from 5-ALA, in tumor tissue is observed with a violet-blue laser light.

quately prevented by avoiding sun exposure for approximately 24 hours after the administration.

Application of 5-ALA in fluorescence-guided glioma surgery

Before induction of general anesthesia, 20 mg/kg of 5-ALA (Cosmo Oil Co., Ltd.) is dissolved in 50 ml of 5% glucose and administered orally. After the induction of general anesthesia, routine craniotomy is conducted. We use a semiconductor laser system (VLDM1, M & M Co., Ltd.) as the excitation light source. During the tumor resection, red fluorescence emitted from the Pp IX in the tumor tissue is observed with a 405-nm violet-blue light emitted from a laser system (Figure 3), which allows normal brain tissue to be differentiated from the tumor tissue.

Intraoperative identification of the margin between a glioma and normal brain tissue is the objective of PDD using 5-ALA. A rondomised cotrolled clinical phase III trial showed that fluorescence-guided surgery with 5-ALA improved progression-free survival in glioma patients.[20] However, the fluorescein may occasionally be weak and autologous fluorescence in the background may also influence the identification. Therefore, more objective and quantitative indicators need to be established. Recently, a quantitative determination method for fluorescence involving spectroscopic analysis has been developed, which allows more precise visualization of the fluorescence from tumor tissue.[22] Regarding the specificity of PDD for glioma surgery, new techniques allowing more specific accumulation of fluorescent dyes in tumors need to be developed. We have found several glioma antigens that are strongly expressed in glioma tissues, but not in the normal brain.[39–43] Fluorescent dyes labeled with antibodies against glioma antigens may be useful to increase the specificity of PDD in glioma surgery.

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