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Endoscopic identification and biopsy sampling of an intraventricular malignant glioma using a 5–aminolevulinic acid–induced protoporphyrin IX fluorescence imaging system

Technical note

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✓Several neurosurgical studies have provided descriptions of the utility of fluorescence-guided tumor resection using a microscope. However, fluorescence-guided endoscopic detection of a deep-seated brain tumor has not yet been reported. The authors report their experience with an endoscopic biopsy procedure for a malignant glioma within the third ventricle using a 5–aminolevulinic acid (5-ALA)–induced protoporphyrin IX fluorescence imaging system. A 5-ALA–induced fluorescence image of an intraventricular tumor is barely visible with the typical fluorescence endoscopic system used in other clinical fields because the intensity of excitation light at wavelengths of 390 to 405 nm through a cutoff filter is too weak to delineate a brain tumor.

The technique described in this study made use of a laser illumination system with a high-powered output that delivered a violet-blue light at wavelengths of 405 nm. In addition, a common ultraviolet cutoff filter was fitted between the endoscope and the high-sensitivity camera to block the backscattered excitation light. A 5-ALA–induced fluorescence endoscopy performed using this system allowed the intraventricular tumor to be clearly visualized as a red fluorescent lesion. Several biopsy specimens obtained from the fluorescent lesion provided a definitive histological diagnosis. The results indicate that this endoscopic system is useful in detecting an intraventricular fluorescent tumor.

Key Words • intraventricular malignant glioma • 5–aminolevulinic acid • biopsy • photodynamic diagnosis • fluorescence-guided resection

In recent years, photodynamic diagnosis and therapy for high-grade malignant gliomas have received increasing attention.[5,6,11–15] Fluorescent dyes that include fluorescein sodium,[6,13] 5-ALA,[11,15] and porfimer sodium[12] are used for photodynamic diagnosis and therapy. A surgical microscope with a fluorescence system can facilitate the resection of malignant gliomas because fluorescent images enable the surgeon to discriminate between the tumor tissue and the surrounding brain tissues.[6,15] In clinical fields other than neurosurgery, several studies have been conducted to investigate the fluorescence endoscopic detection of premalignant and malignant lesions.[1,4,7,10,16] However, there has been no report of a photodynamic diagnosis of a deep-seated brain tumor using fluorescence endoscopy, primarily because the fluorescence intensity under ordinary excitation light is too weak to allow observation of the central nervous system. Therefore, we developed a 5-ALA–induced fluorescence endoscopic system equipped with a high-powered laser light source. In this report we describe the clinical usefulness of a fluorescence endoscopic detection and biopsy procedure for an intraventricular malignant glioma.

Operative Technique

A flexible fiberscope with an external diameter of 4.2 mm (N-4L, Machida) was used for the procedure. The fiberscope was connected to either a light source delivering common white light (Olympus) or to a laser illumination system delivering ultraviolet laser light (BP-300, Ball Semiconductor, Inc.). This laser system was equipped with a bundle of 16 optical fibers emitting a 405 nm wavelength laser and a 300-mW high-powered output. An ultraviolet cutoff filter (Wratten No. 2E, Kodak) was inserted between the ocular lens of the fiberscope and the high-sensitivity 2D charge-coupled device camera (Hamamatsu Photo) to cut off reflective light, including excitation light. This filter does not influence the normal color tone of the image, so it

Abbreviations used in this paper: BBB = blood–brain barrier; CSF = cerebrospinal fluid; 5-ALA = 5–aminolevulinic acid.

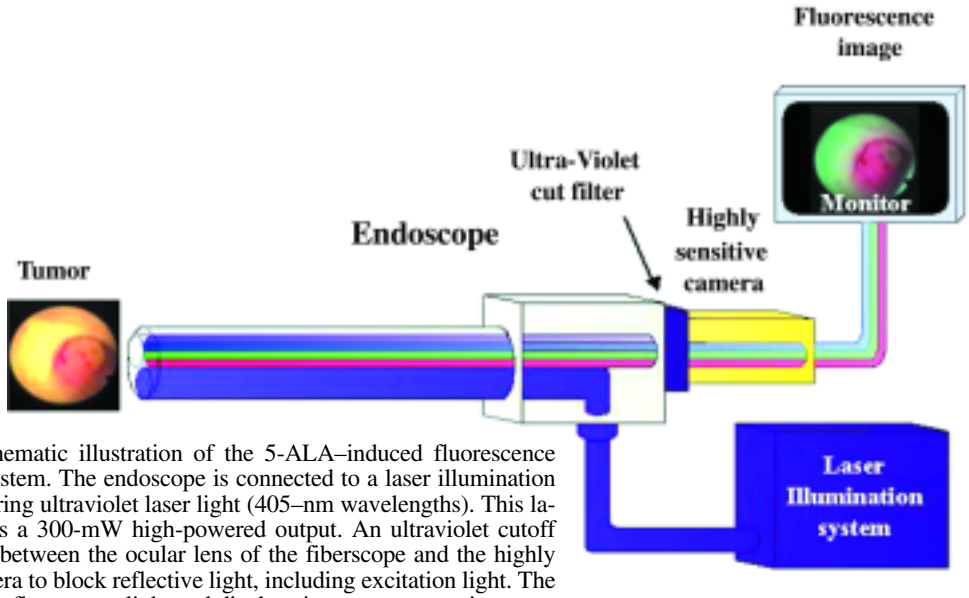

FIG. 1. Schematic illustration of the 5-ALA–induced fluorescence endoscopic system. The endoscope is connected to a laser illumination system delivering ultraviolet laser light (405–nm wavelengths). This laser system has a 300-mW high-powered output. An ultraviolet cutoff filter is fitted between the ocular lens of the fiberscope and the highly sensitive camera to block reflective light, including excitation light. The camera detects fluorescent light and displays images on a monitor.

does not need to be changed between the common white light and laser light modes. The camera was used to capture fluorescence images by gathering background autofluorescence in normal brain tissue and 630 nm wavelength red fluorescence in the malignant tumor; the images were then depicted on a monitor (Fig. 1).

Illustrative Case

History and Examination. This 56-year-old woman presented with a 1-month history of progressive headaches and diplopia. Magnetic resonance images demonstrated hydrocephalus due to a nonenhancing midbrain mass lesion. The patient underwent an endoscopic tumor biopsy procedure and third ventriculostomy at a nearby hospital. Her histological examination revealed a possible low-grade glioma because the removed specimens were small. She did not receive adjuvant therapy for this tumor. However, 6 months later she again presented with diplopia. Additional magnetic resonance images showed a ring-enhancing lesion in the same midbrain area (Fig. 2). The patient was referred to our hospital for a definitive diagnosis and further treatment.

Fluorescence Endoscopic Biopsy Procedure. We obtained approval to perform the fluorescence endoscopic biopsy procedure from the Ethical Committees of Osaka Medical College. Four hours before surgery, the patient received 20 mg/kg of 5-ALA (Medac GmbH) dissolved in water and administered orally. After anesthesia was induced, the right lateral ventricle was cannulated with a No. 16 peelaway catheter. The fiberscope was inserted into the third ventricle through the foramen of Monro, and a good CSF flow through the previous stoma was identified by floor pulsation. The endoscope was passed farther into the posterior third ventricle and a protruding lesion was visualized at the entrance of the aqueduct. Under white light, this lesion was similar in color to the surrounding brain tissue (Fig. 3A). When the light source of the endoscope was changed to the laser light, the lesion emitted a red fluorescence (Fig. 3B). Using small forceps, several biopsy samples were taken from this red fluorescent lesion.

Postoperative Course. The histological diagnosis of the biopsy specimens was consistent with anaplastic astrocytoma (Fig. 4). The patient received local radiotherapy totaling 60 Gy and chemotherapy with nitrosourea. Her visual symptoms responded well to these therapies, and she was able to walk without aid.

Discussion

We previously used fluorescein sodium as a fluorescent dye in the microscopic photodynamic diagnosis of malignant gliomas.[5,6] Fluorescein sodium passes into the brain tissue and brain tumor through the vessels, including the capillaries, where the BBB is absent or disrupted. Following intravenous administration of fluorescein sodium, all of the tissues without a BBB emit green fluorescence. We have

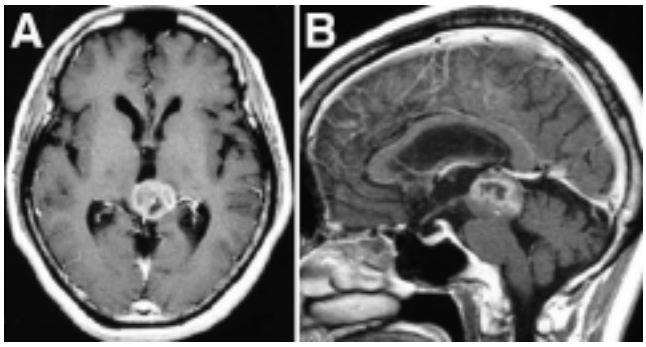

FIG. 2. Axial (A) and sagittal (B) T_1-weighted magnetic resonance images with contrast showing a heterogeneous enhanced mass in the posterior portion of the third ventricle.

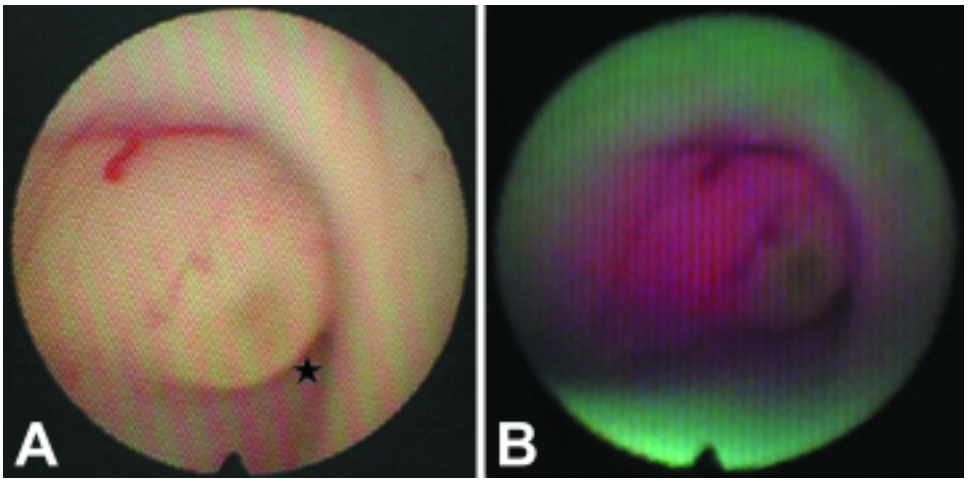

FIG. 3. Endoscopic photographs of the entrance of the aqueduct *(star)*. Compared with a typical endoscopic image system (A), the 5-ALA–induced fluorescence endoscopic system shows a red fluorescent mass (B).

often been unable to discriminate between tumor tissues and peritumoral vasogenic edema using the fluorescence microscopic system. Fluorescein sodium was not suitable for intraventricular tumor detection by the endoscope because it rapidly diffuses into the CSF.

In contrast, 5-ALA is absorbed into both normal and tumor tissues, whereas the metabolite protoporphyrin IX accumulates selectively in the tumor tissue. This difference is probably due to altered activity levels of the enzymes of the heme biosynthetic pathway within the transformed cells.[3,16] Only protoporphyrin IX within the tumor emits red fluorescence under violet-blue light; therefore, 5-ALA is a promising fluorescent dye to use in inspection of the CSF space.

In neurosurgery, 5-ALA–induced fluorescence has been used only in conjunction with a surgical microscope. Stummer and colleagues[14] reported the usefulness of fluorescence-guided resection with 5-ALA–induced porphyrins for glioblastomas. However, there has not been a report on efforts to detect a brain tumor under an endoscopic fluorescence image using 5-ALA. In the oropharyngolarynx, bladder, uterus, gastrointestinal system, and other areas within the body that do not have a barrier such as the BBB, 5-ALA can easily move from the blood vessels to the neoplasm. Therefore, protoporphyrin IX emits relatively stable fluorescence, and it is possible to detect the fluorescent lesion using fluorescence endoscopy.[1,4,7,10,16]

In the conventional fluorescence endoscopic system, a 300-mW short-arc xenon light source is used. This instrument is equipped with a dielectric short-pass filter (375–440 nm wavelengths), which exchanges a white light for a violet-blue light so that the intensity of the excitation light is decreased. For this reason, it was difficult to detect a red fluorescent intraventricular tumor by the ordinary system (unpublished data). In the present endoscopic system, we provided a high-powered laser illumination system as a light source. This 405 nm–wavelength laser light delivers an excitation light that ensures an adequate 5-ALA–induced fluorescence intensity in the central nervous system. Because an ultraviolet cutoff filter that blocked the backscattered excitation laser light was inserted between the endoscope and the camera, the intraventricular structure and the tumor were visible under both the white light and the laser light without having to change the filter. This system is simple and economical. Therefore, its clinical application is promising for fluorescence endoscopic detection of malignant brain tumors.

Due to the limited penetration depth of protoporphyrin IX,[8] it is, however, difficult to use a fluorescence image to detect a tumor that is covered with brain tissue, except for the thin ependymal layer. If technical advances in fluorescence images are able to solve this problem, it would be possible to locate not only intraventricular but also intraparenchymal tumors. Moreover, if the ongoing photodynamic therapy research on malignant tumors[2,9,11,12] becomes established in the foreseeable future, we can speculate that the fluorescence endoscopic system has a good chance of

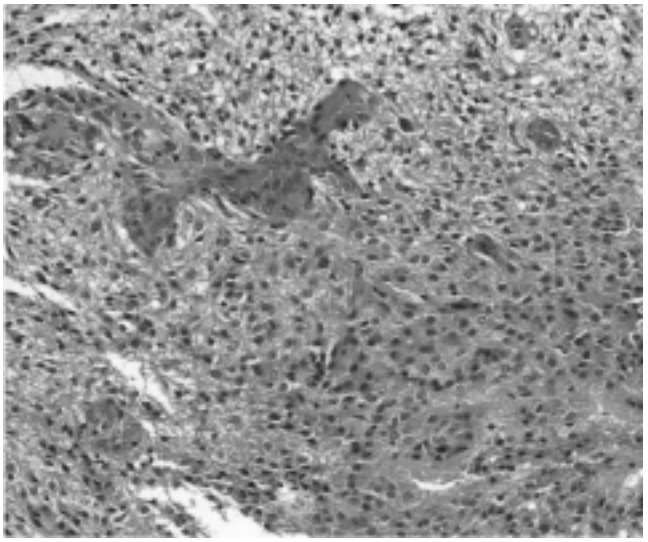

FIG. 4. Photomicrograph of tumor tissue obtained from the red fluorescent area in Fig. 3 showing high cellularity, pleomorphism, and endothelial proliferation. The tumor was later diagnosed as an anaplastic astrocytoma. H & E, original magnification × 100.

becoming the preferred treatment for deep-seated brain tumors.

Conclusions

We developed a simple and economical fluorescence endoscopic system that was very useful for intraoperative detection and diagnosis of malignant gliomas in the third ventricle. We expect that this system will form the basis for fluorescence endoscopic neurosurgery in the future.

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