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Simple Summary: This study investigated the effectiveness of combining intraoperative ultrasonography (IOUS) with sodium fluorescein (SF) to evaluate tumor resection completeness in glioblastoma isocitrate dehydrogenase-1 (IDH1)-wildtype patients. The goal was to compare this approach to postoperative magnetic resonance imaging (MRI) for detecting residual tumors. The study included 97 patients who underwent surgery between 2015 and 2024. One group received SF alone, and the other received SF with IOUS. The group using SF with IOUS had a higher gross total resection (GTR) rate (83.3%) than the SF-only group (67.3%), but the difference wasn't statistically significant. Postoperative MRI showed more residual tumors in the SF group (12.2%) compared to the SF with IOUS group (8.3%). The survival rate was significantly higher in the SF with IOUS group (24 months) compared to the SF-only group (14 months). However, there were no significant differences in new neurological deficits or patient performance scores between the groups post-surgery. In conclusion, SF combined with IOUS is a reliable method for more complete tumor resection and better surgical outcomes, though further research is needed to refine its use in neurosurgery.

Abstract: Background/Objectives: This study investigated the effectiveness of intraoperative ultrasonography (IOUS) combined with sodium fluorescein (SF) in evaluating tumor resection completeness in patients with glioblastoma IDH1-wildtype. By comparing SF with IOUS with postoperative magnetic resonance imaging (MRI) for detecting residual tumors, we aimed to evaluate its potential in improving surgical precision and neurosurgical outcomes. **Methods**: Adult patients with supratentorial IDH-wildtype grade 4 glioblastoma who underwent resection using SF or SF with IOUS during 2015–2024 were included. **Results**: A total of 97 patients met the inclusion criteria (49 SF group and 48 SF with IOUS group). The gross total resection (GTR) rate was higher in the SF with IOUS group (83.3%) than in the SF group (67.3%), although the difference was not statistically significant (p = 0.112). For residual tumors according to postoperative MRI findings as a result of subtotal resection due to tumor invasion of eloquent anatomical locations, 6/49 (12.2%) patients in the SF group showed a positive result ($\varkappa$: 0.447, p = 0.001), and 4/48 (8.3%) patients in the SF with IOUS group showed a positive result ($\varkappa$: 0.625, p < 0.001). The sensitivity, specificity, negative predictive value, and positive predictive value for predicting residual tumors peroperatively compared with postoperative MRI results were calculated for the SF and SF with IOUS groups. Comparison between the SF and SF with IOUS groups revealed a statistically significant difference in the estimated mean survival time, with 14 months (standard error: 1.236) for the SF group and 24 months (standard error: 4.103) for the SF with IOUS group (p < 0.001). In total, 11/49 (22.4%) patients in the SF group, and 10/48 (20.8%) patients in the SF with IOUS group experienced newly developed neurological deficits postoperatively (p > 0.05). In the SF with IOUS group, 26/48 (54.2%) patients,

14/48 (29.2%) patients, and 8/48 (16.7%) patients had Karnofsky Performance Status scores of 90–100, 70–80, and <70, respectively (p = 0.525), and 12 patients experienced deterioration, 24 patients were stable, and 12 patients had improved at 1 month. **Conclusions**: SF with IOUS provides a reliable imaging modality for achieving successful GTR and improving surgical outcomes. Nevertheless, further research is necessary to overcome its limitations and better define its intraoperative role.

Keywords: fluorescein; ultrasound; resection; glioblastoma; surgery

1. Introduction

Advancements in neurosurgical oncology have continuously driven the development of novel technologies designed to improve surgical precision and patient outcomes. Among these innovations, intraoperative neuroimaging has evolved significantly, allowing for more extensive tumor resections and minimizing neurological risks [1–4]. In various brain tumor cases, the extent of tumor removal is closely associated with improved survival rates and responses to adjuvant therapies [5–10]. Therefore, refining surgical techniques to maximize tumor excision remains a key objective in neurosurgery [8].

Various intraoperative imaging modalities are available to neurosurgeons, including intraoperative magnetic resonance imaging (IOMRI), intraoperative computed tomography (IOCT), intraoperative ultrasonography (IOUS), and fluorescence-guided methods such as sodium fluorescein (SF) and 5-aminolevulinic acid (5-ALA) [2,4]. These imaging techniques are frequently integrated into standard neurosurgical workflows and neuronavigation systems [4]. Each modality presents distinct advantages and limitations, and there is no universally accepted gold standard [11–14]. The selection of an imaging approach is typically influenced by a combination of surgeon preference and institutional resources [4]. A primary challenge in achieving maximal resection is encountered in cases where tumor boundaries are poorly defined, making it difficult to differentiate tumor tissue from surrounding brain structures [2].

Gliomas are the most common primary brain tumors in adults, with an estimated incidence of 3–5 cases per 100,000 people annually [15–19]. Although these tumors can develop at any age and in various regions of the central nervous system, they predominantly occur intracranially in individuals aged 50–60 years [16]. The World Health Organization classifies gliomas on the basis of their cellular origin—such as astrocytic or oligodendroglial—and histological grade [20]. Among these, high-grade gliomas (HGGs), including grade 3 and 4 tumors, are particularly aggressive and associated with poor prognoses [19].

Despite significant advancements in neurosurgical techniques and ongoing research, glioblastomas and other HGGs remain highly lethal, with the 5-year survival rates still <5% [2–5]. Total gross resection (GTR) has been identified as a crucial factor in improving both progression-free survival (PFS) and overall survival (OS). However, complete tumor removal is often unachievable in cases where tumors are adjacent to or involve critical brain structures, because aggressive resection in these regions could result in severe functional impairment [21].

Among intraoperative imaging techniques, IOUS has gained recognition as a valuable tool for real-time tumor localization and resection assessment [22–24]. A key advantage of IOUS is its ability to provide dynamic intraoperative visualization, enabling surgeons to adjust their approach during surgery with greater accuracy [23–25]. Compared with IOMRI and IOCT, IOUS provides remarkable practical benefits, including lower cost, ease of use, and wider accessibility, and also demonstrates effectiveness in identifying specific tumor characteristics [23,26–28]. Due to these advantages, IOUS has been successfully implemented in both adult and pediatric populations as a supplementary imaging tool to improve the delineation of tumor margin and the extent of resection [23,24,27,29,30].

The present study, in addition to our previous research conducted using SF [31], aims to further explore the effectiveness of IOUS, in addition to SF, in evaluating the completeness of tumor resection

and the impact of using SF alone versus SF combined with IOUS on surgical resection and patient survival. Based on existing research, we anticipate that IOUS, when used along with SF, will significantly contribute to real-time surgical decision-making, enable more precise tumor resection, and improve clinical outcomes in neurosurgical patients.

2. Materials and Methods

2.1. Patient Characteristics and Data Collection

This retrospective study evaluated adult patients (aged ≥18 years) diagnosed with supratentorial IDH-wildtype grade 4 glioblastoma, through histopathological examination, who underwent surgical resection during 2015–2024 at the Neurosurgery Department of Kocaeli University Faculty of Medicine and the Neurosurgery Department of Derince State Hospital, Turkey. Demographic, clinical, radiological, and surgical data were retrospectively collected for these patients. The collected variables included patient age at the time of surgery, date of operation, preoperative and intraoperative imaging findings (SF, SF with IOUS, and MRI), tumor location, operative records, and postoperative imaging assessments. The presence of residual tumor was determined using intraoperative findings and postoperative MRI, as evaluated by an experienced neuroradiologist. Follow-up physician records and MRI scans conducted within 3 months postsurgery were also reviewed.

Patients included in the study underwent surgery using either SF alone or a combination of SF and IOUS for intraoperative tumor visualization. GTR was preoperatively defined as the complete removal of all T1-weighted contrast-enhancing tumor tissues on MRI. Surgical interventions were performed by two experienced neurosurgeons.

The exclusion criteria included incomplete medical records, procedures limited to intracranial biopsy or planned subtotal tumor debulking, and tumors located in deep eloquent structures such as the basal ganglia, thalamus, and brainstem. These locations were excluded because of the inherent difficulty in achieving GTR and preserving neurological function.

2.2. Surgical Procedure

Patients were categorized into two groups based on the intraoperative imaging techniques used; one group underwent resection with SF alone, and the other group underwent a combination of SF and IOUS. Patients were excluded if they did not receive one of these two imaging approaches or if available data were insufficient to confirm intraoperative imaging use. Throughout all procedures, efforts were made to preserve eloquent brain structures and minimize neurological deficits. Whenever feasible, GTR was pursued. Intraoperative neurophysiological monitoring, including motor and somatosensory assessments, was used along with neuronavigation systems.

Preoperative MRI with and without contrast was conducted for all patients to assist in surgical planning. In the operating room, patients were positioned in a Mayfield head holder for stabilization and optimal surgical access. Neuronavigation was used in both groups for intraoperative guidance.

In the SF group, a 5 mg/kg intravenous dose of SF was administered after anesthesia induction, according to established protocols [32]. Fluorescence-guided resection was performed using a surgical microscope equipped with xenon white light illumination. Initial fluorescence was observed throughout the brain parenchyma; however, over time, it remained concentrated in areas with blood–brain barrier (BBB) disruption. Resection was guided by fluorescence-stained tissue, indicating tumor regions that required removal. Resection was continued until no fluorescence remained, suggesting the transition to normal brain parenchyma. Tumors were primarily excised en bloc, although piecemeal removal using an ultrasonic aspirator was utilized in some cases (Figure 1).
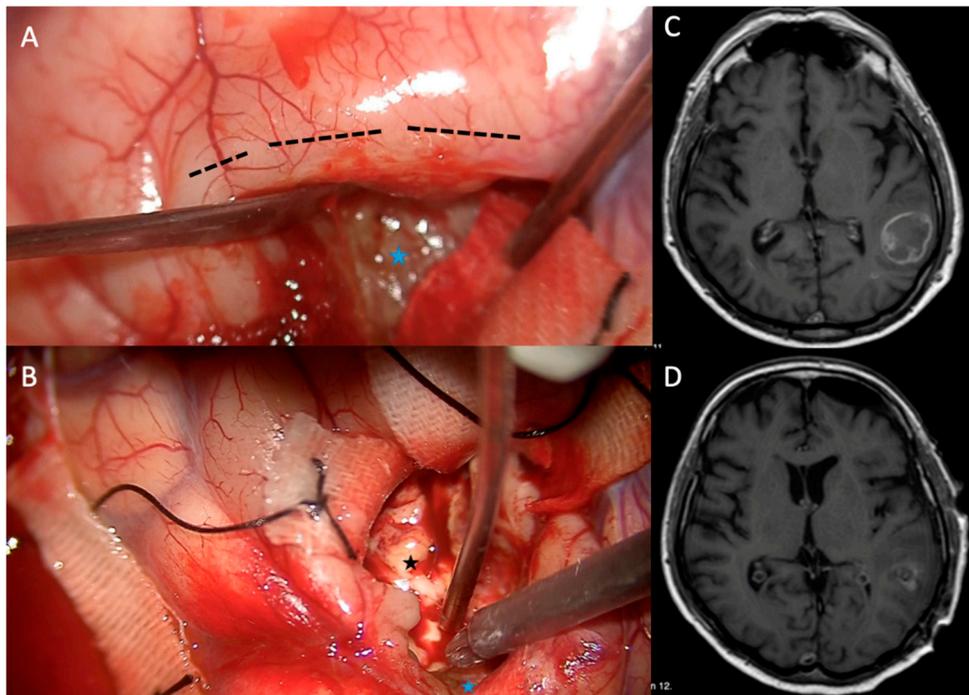
Figure 1. A. The tumor tissue stained with SF was reached after sulcal dissection. **B.** After tumor resection, the underlying edematous normal brain parenchyma was observed, and resection of the remaining tumor tissue stained with SF continued using an ultrasonic aspirator. **C and D.** Preoperative and postoperative contrast-enhanced MRI views. Black dashed line shows gyrus. Black asterisk shows edematous normal brain parenchyma after resection. Blue asterisk shows remaining tumor tissue stained with SF.

In the SF with IOUS group, SF was administered as described earlier, and IOUS was used concurrently to examine the tumor cavity for residual tissue. A standard two-dimensional ultrasound probe (LOGIQ Ultrasound, GE Healthcare) was used at various stages to optimize resection. In both groups, a surgical microscope with xenon white light illumination was used, without additional filters or modifications.

A craniotomy was performed to expose the dura, and sterile saline irrigation was applied to the surgical field to improve ultrasound transmission. Neuronavigation and IOUS were performed to confirm tumor localization and delineate margins. IOUS was periodically performed throughout the resection to evaluate progress. When the tumor was believed to be completely resected, a final IOUS scan was performed. If a residual tumor was detected in an area considered safe for further resection, additional excision was attempted. However, if the tumor was located in an eloquent region where further resection posed a high risk of neurological compromise, intentional subtotal resection was performed to preserve function. The IOUS findings were interpreted in real-time by the attending neurosurgeon to guide intraoperative decision-making (Figure 2).
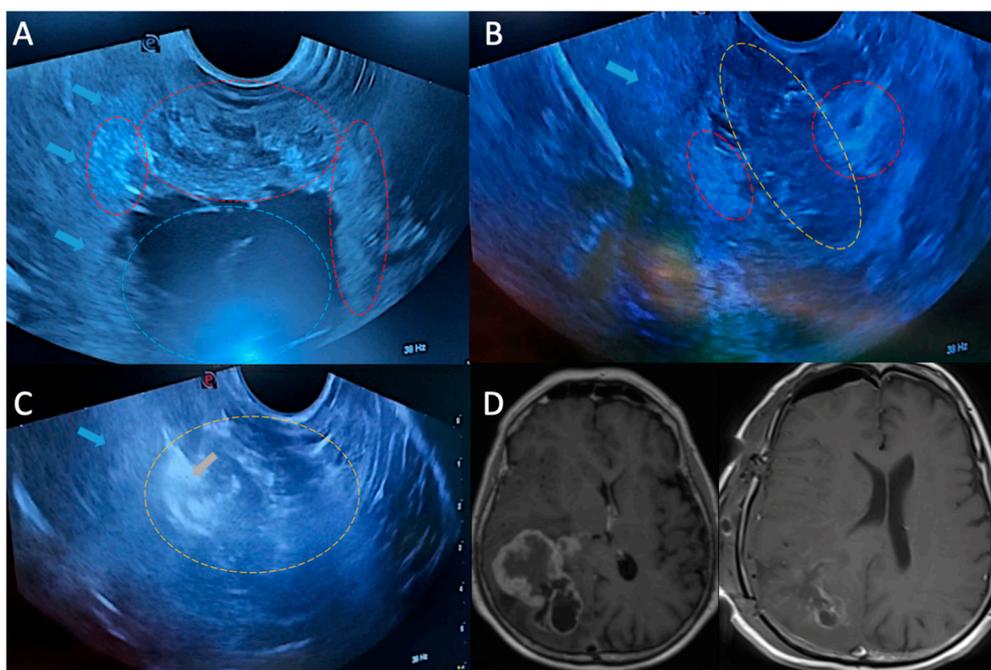
Figure 2. A. IOUS were performed to confirm tumor localization and delineate margins. Red dashed lines indicate the solid tumor portion, blue dashed line indicate the cystic part of the tumor. **B.** IOUS was performed throughout the resection to evaluate progress. In the IOUS, red dashed lines indicate the remaining solid tumor mass, yellow dashed line shows the resection cavity. **C.** Post-resection IOUS image. Yellow dashed line indicates the GTR surgical cavity, and gray arrow shows the hemostatic agents placed in the cavity. **D.** Preoperative and postoperative contrast-enhanced MRI views. Blue arrows show the edematous normal brain tissue surrounding the tumor.

At the completion of resection, the primary intraoperative imaging modality—either SF alone or SF with IOUS—was used to examine the resection cavity for residual tumor. Postoperative MRI was considered the gold standard for confirming residual contrast-enhancing tissue. If residual tumor was detected on intraoperative imaging and further resection was deemed safe, additional excision was performed until GTR was achieved or further removal was considered high risk due to proximity to critical neurovascular structures.

2.3. Surgical Outcomes

The primary outcome measures included the extent of tumor resection, presence of residual tumor on postoperative imaging, achievement of GTR, accuracy of intraoperative imaging in detecting residual tumor, and the incidence of new postoperative neurological deficits. The extent of resection was quantified as the percentage reduction in tumor volume between preoperative and postoperative MRI scans. Residual tumor was defined as any contrast-enhancing lesion detected on postoperative MRI, as evaluated by an attending neuroradiologist. GTR was defined as the complete removal of the contrast-enhancing tumor tissue, confirmed on T1-weighted MRI with gadolinium contrast.

False-negative intraoperative assessments were identified in cases where intraoperative imaging indicated complete resection, but postoperative MRI revealed residual tumor. The development of new neurological deficits was evaluated during the initial follow-up visit, approximately 2 weeks postsurgery. Any new or worsening deficits were considered indicators of possible overresection based on intraoperative imaging guidance.

2.4. Radiographic Assessment and Follow-Up

Preoperative and postoperative MRI scans were evaluated to establish baseline tumor size and determine the extent of resection. T1-weighted contrast-enhanced sequences were used for evaluating imaging findings, with only the surgically treated lesion considered in cases with multiple contrast-enhancing foci. An experienced neuroradiologist, blinded to patient outcomes, reviewed all the imaging findings.

Routine follow-up MRI examinations were conducted at regular intervals, with shorter intervals applied in cases where findings were ambiguous or where disease progression was suspected.

2.5. Measures of Diagnostic Accuracy

At the end of surgery, SF or SF with IOUS was used to evaluate the resection cavity for any remaining tumor. Findings were compared against postoperative gadolinium-enhanced MRI, which served as the reference standard for detecting residual tumor. Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were calculated for both SF alone and SF with IOUS groups to evaluate their intraoperative diagnostic accuracy.

2.6. Survival and Functional Outcomes

OS was defined as the time from primary tumor resection to death. PFS was determined as the interval from the initial resection to malignant progression, confirmed by either histopathological findings from reoperation or new contrast-enhancing lesions on follow-up MRI.

Functional status was evaluated using the Karnofsky Performance Status (KPS) scale, which was recorded both preoperatively and 1 month postoperatively. The KPS scale score ranges from 100 (normal function) to 10 (moribund state), with higher scores indicating better survival and quality of life.

2.7. Statistical Analysis

All statistical analyses were conducted using IBM SPSS 29.0 (IBM Corp., Armonk, NY, USA). Kolmogorov–Smirnov and Shapiro–Wilk tests were conducted to evaluate the normality assumption. Continuous variables were expressed as mean ± standard deviation or median and interquartile range (IQR). Categorical variables were summarized as counts and percentages. Comparisons between groups were conducted using the independent samples t-test for normally distributed variables and the Mann–Whitney U test for nonnormally distributed variables. Associations between categorical variables were analyzed using the chi-square test. The Kaplan–Meier method with the log-rank test was used for survival analysis. Agreement analysis was conducted using Kappa's statistic. $p < 0.05$ was considered statistically significant.

3. Results

3.1. Characteristics of Patients

Table 1 shows the characteristics of 97 patients (49 in the SF group and 48 in the SF with IOUS group) who were treated for IDH-wildtype grade 4 glioblastoma.

The male/female proportion of patients in the SF group was 22/27, and that in the SF with IOUS group was 30/18 (SF: 44.9%; IOUS: 62.5%; p = 0.125). The mean age of patients was 53.04 ± 8.21 years in the SF group and 57.29 ± 9.74 years in the SF with IOUS group (p = 0.022).

Table 1. Characteristics, clinical presentation and preoperative neuroimaging findings of patients. Quantitative variables are expressed as mean ± standard deviation (range), and qualitative variables are expressed as *n* (%). IOUS: intraoperative ultrasonography, SF: sodium fluorescein, NA: Not applicable.

	SF Group (n=49)	SF with IOUS Group (n=48)	pValue
Sex			
Male	22 (44.9%)	30 (62.5%)	0.125
Female	27 (55.1%)	18 (37.5%)	
Age	53.04 ± 8.21 (34-70)	57.29 ± 9.74 (38-79)	0.022
Comorbidities			
Yes	17 (34.7%)	22 (45.8%)	0.362
No	32 (65.3%)	26 (54.2%)	
History of Operation			
Yes	13 (26.5%)	14 (29.2%)	0.950
No	36 (73.5%)	34 (70.8%)	
Presenting Symptom			
Headache (n=51)	21 (41.2%)	30 (58.8%)	0.083
Seizure (n=31)	17 (54.8%)	14 (45.2%)	0.714
Paresis (n=21)	11 (52.4%)	10 (47.6%)	1.000
Speech Impairment (n=14)	8 (57.1%)	6 (42.9%)	0.805
Cognitive Changes (n=11)	6 (54.5%)	5 (45.5%)	1.000
	0	4 (100%)	NA
Visual Disturbances (n=4)	1 (33.3%)	2 (66.6%)	NA
Incidental (n=3)			
Preoperative KPS Skore			
90-100	23 (46.9%)	20 (41.7%)	
70-80	22 (44.9%)	22 (45.8%)	0.733
<70	4 (8.2%)	6 (12.5%)	
Maximal Diameter of Tumor	46.94 ± 9.44 (29-68)	46.15 ± 8.98 (32-64)	0.605

3.2. Clinical Presentation

The preoperative clinical features are also summarized in Table 1. Comorbidities were detected in 17 patients (34.7%) in the SF group and 22 patients (45.8%) in the SF with IOUS group (p = 0.362). Recurrent tumors were more frequent in the SF with IOUS group than in the SF group. A total of 36 patients (73.5%) in the SF group and 34 patients (70.8%) in the SF with IOUS group had no previous history of operation (p = 0.950) (The remaining 13 (26.5%) vs 14 (29.2%) patients had undergone previous surgical procedures). The most common presenting symptoms in both groups were headache in 51/97 (52.6%) patients, seizure in 31/97 (32%) patients, paresis in 21/97 (21.6%) patients, speech impairment in 14/97 (14.4%) patients, cognitive changes in 11/97 (11.3%) patients, and visual disturbances in 4/93 (4.1%) patients.

In the SF group, 23/49 (46.9%) patients, 22/49 (44.9%) patients, and 4/49 (8.2%) patients had a preoperative KPS score of 90–100, 70–80, and <70, respectively. In the SF with IOUS group, 20/48

(41.7%) patients, 22/48 (45.8%) patients, and 6/48 (12.5%) patients had a preoperative KPS score of 90–100, 70–80, and <70, respectively (p = 0.733).

3.3. Preoperative Neuroimaging Findings and Tumor Features

The tumor characteristics and neuroimaging findings are shown in Table 1. The most common tumor localizations were left frontal (11/49), left temporal (7/49), and left parietal (6/49) in the SF group and left frontal (8/48), left parietal (8/48), and right temporal (8/48) in the SF with IOUS group. Preoperative MRI measurements revealed a mean ± standard deviation (range) maximal diameter of the tumor of 46.94 ± 9.44 (29–68) mm in the SF group and 46.15 ± 8.98 (32–64) mm in the SF with IOUS group (p = 0.605).

3.4. Surgical Evaluation and Tumor Resection Outcomes

Table 2 shows the outcomes of patients' surgical evaluation and tumor resection.

In total, 33/49 (67.3%) patients in the SF group and 40/48 (83.3%) patients in the SF with IOUS group had a negative result (GTR) for residual tumor according to postoperative MRI. The GTR rate was higher in the SF with IOUS group (83.3%) than in the SF group (67.3%), although the difference was not statistically significant (p = 0.112).

A total of 6/49 (12.2%) patients in the SF group ($\varkappa$: 0.447, p = 0.001) and 4/48 (8.3%) patients in the SF with IOUS group ($\varkappa$: 0.625, p < 0.001) had a positive result for residual tumor according to both preoperative techniques and postoperative MRI as a result of subtotal resection (STR) due to tumor invasion of eloquent anatomical locations. For detecting residual tumor, the agreement analysis was moderate in the SF group but substantial in the SF with IOUS group, according to Kappa's statistic.

Residual tumor could be correctly identified using SF in 6/16 (37.5%) STR cases, whereas using SF with IOUS, it could be correctly identified in 4/8 (50%) STR cases, according to postoperative MRI.

Among patients with recurrent tumors, 5/13 in the SF group and 2/14 in the SF with IOUS group had false-negative results. All patients with recurrent tumors had undergone surgery, chemotherapy, and radiotherapy previously.

Table 2. Patients surgical evaluation and tumor resection outcomes. Qualitative variables are expressed as *n* (%).(%). IOUS: intraoperative ultrasonography, SF: sodium fluorescein, MRI: Magnetic resonance imaging.

	SF Group (n=49)		SF with IOUS Group (n=48)	
	No Residual Tumor	Residual Tumor	No Residual Tumor	Residual Tumor
Postoperative MRI				
No Residual Tumor	30 (61.2%)	3 (6.1%)	36 (75%)	4 (8.3%)
Residual Tumor	10 (20.4%)	6 (12.2%)	4 (8.3%)	4 (8.3%)
	Recurrent SF Group (n=13)		Recurrent SF with IOUS Group (n=14)	
	No Residual Tumor	Residual Tumor	No Residual Tumor	Residual Tumor
Postoperative MRI				
No Residual Tumor	4 (30.8%)	3 (23%)	6 (42.8%)	4 (28.6%)
Residual Tumor	5 (38.5%)	1 (7.7%)	2 (14.3%)	2 (14.3%)

3.5. Measures of Diagnostic Accuracy

Table 3 shows the sensitivity, specificity, NPV, and PPV for predicting residual tumor preoperatively compared with postoperative MRI results in the SF and SF with IOUS groups. The

respective values were 37.5% (95% CI 18.5%–61.4%), 100% (95% CI 89.6%–100%), 76.7% (95% CI 62.1%–87.1%), and 100 % (95% CI 90.9%–99.8%) in the SF group and 50% (95% CI 21.5%–78.5%), 100% (95% CI 91.2%–100%), 90.9% (95% CI 78.2%–96.9%), and 100% (95% CI 90.8%–99.8%) in the SF with IOUS group.

Table 3. Comparison of sensitivity, specificity, positive predictive value, and negative predictive value between groups. IOUS: intraoperative ultrasonography, SF: sodium fluorescein, NPV: Negative predictive value, PPV: Positive predictive value, GTR: Gross total resection, CI: Confidence interval.

|	SF Group (n=49)	SF with IOUS Group (n=48)
Sensitivity	0.375 (18.5%–61.4%)	0.500 (21.5%–78.5%)
Specificity	1.000 (89.6%–100%)	1.000 (91.2%–100%)
NPV	0.767 (62.1%–87.1%)	0.909 (CI 78.2%–96.9%)
PPV	1.000 (CI 90.9%–99.8%)	1.000 (90.8%–99.8%)
GTR	33 (67.3%)	40 (83.3%)

3.6. Survival Outcomes

For the entire population, the estimated 1-year OS rate was 71.6%, and the estimated 2-year OS rate decreased to 15%. The estimated mean survival time after surgery for the entire population was 17 months (standard error: 0.686). A statistically significant difference was found in the estimated mean survival time when the SF and SF with IOUS groups were compared, with 14 months (standard error: 1.236) for the SF group and 24 months (standard error: 4.103) for the SF with IOUS group (p < 0.001) (Figure 3).
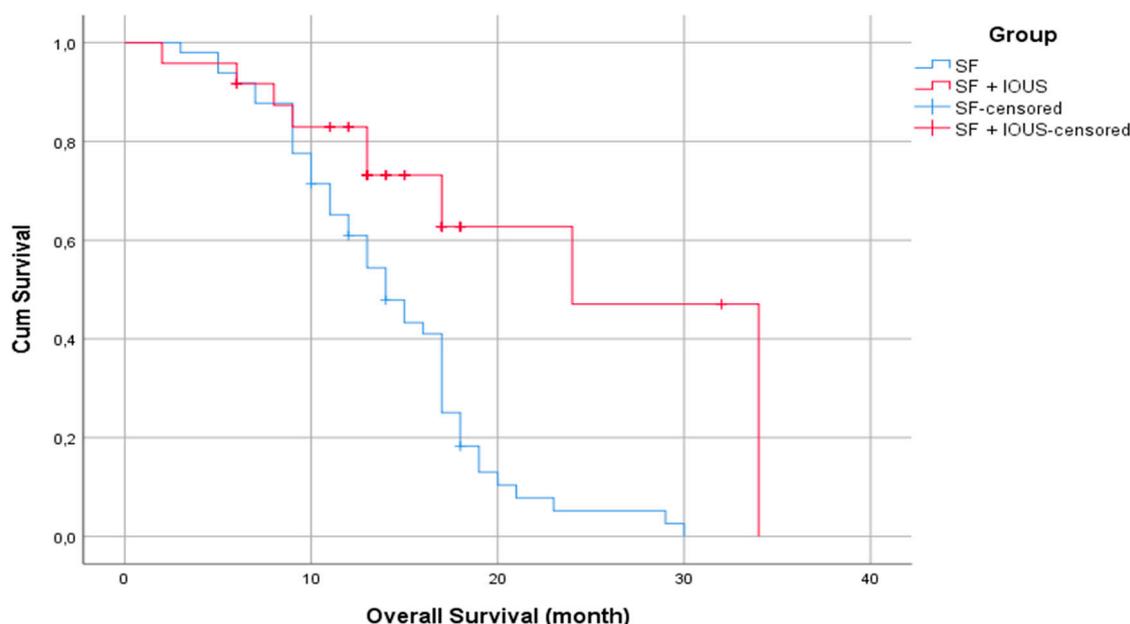
Figure 3. Kaplan–Meier curves for estimated mean survival time (months) between the SF and SF with IOUS groups.

The 1-year PFS rate for the entire population was 47.5%, with a mean estimated PFS of 12 months (standard error: 0.690). However, there was no statistically significant difference in PFS durations between the two groups, with a mean PFS of 12 months (standard error: 1.456) for the SF group and 13 months (0.992) for the SF with IOUS group (p = 0.182) (Figure 4).
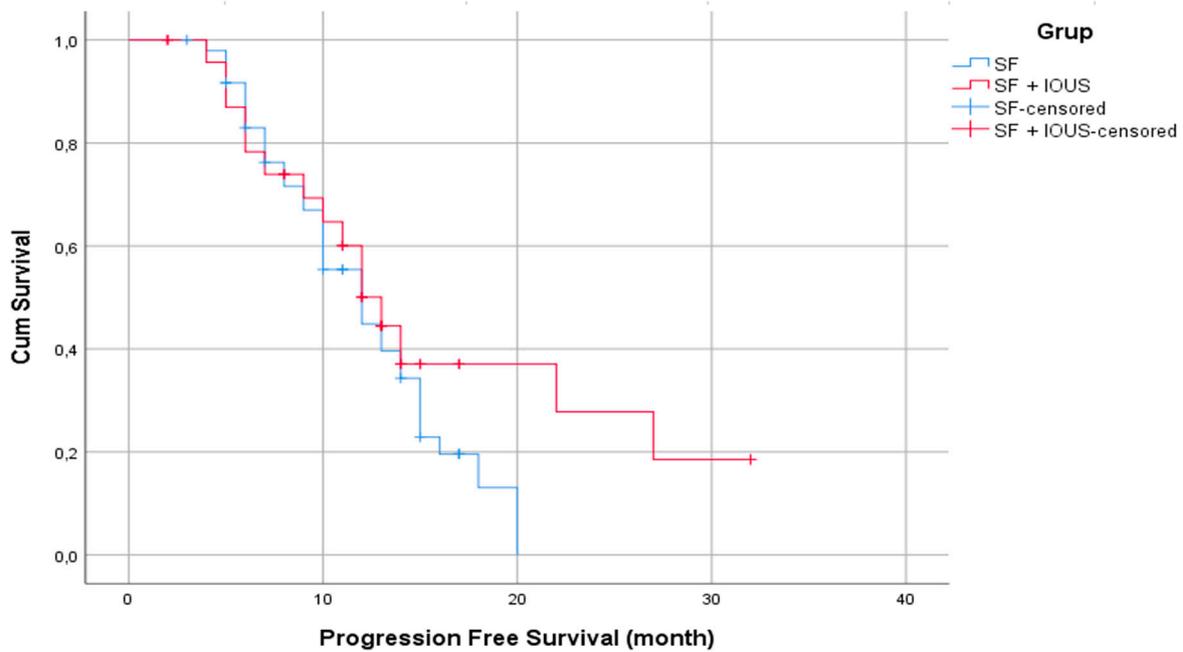
Figure 4. Kaplan–Meier curves for PFS durations (months) between the SF and SF with IOUS groups.

3.7. Postoperative New Neurological Deficits and KPS Scores

Table 4 presents the newly developed postoperative neurological deficits and KPS scores.

Newly developed neurological deficits postoperatively were identified in 11/49 (22.4%) patients in the SF group (1 motor and 1 cognitive deficits were permanent) and 10/48 (20.8%) patients in the SF with IOUS group (1 motor deficit was permanent) (p = 1.000).

In the SF group, 21/49 (42.9%) patients, 19/49 (38.8%) patients, and 9/49 (18.4%) patients had a postoperative KPS score of 90–100, 70–80, and <70, respectively. In total, 14/49 patients experienced deterioration, 26/49 patients were stable, and 9/49 patients had improved at 1 month.

In the SF with IOUS group, 26/48 (54.2%) patients, 14/48 (29.2%) patients, and 8/48 (16.7%) patients had a postoperative KPS score of 90–100, 70–80, and <70, respectively (p = 0.525). A total of 12 patients experienced deterioration, 24 patients were stable, and 12 patients had improved at 1 month.

Table 4. Postoperative newly developed neurological deficits and Karnofsky Performance Status (KPS) score of patients. Qualitative variables are expressed as *n* (%).. IOUS: intraoperative ultrasonography, SF: sodium fluorescein, KPS: Karnofsky Performance Status.

	SF Group (n=49)	SF with IOUS Group (n=48)	p Value
New Neurologic Deficits			
Transient	9 (18.4%)	9 (18.7%)	1.000
Permanent	2 (4.1%)	1 (2.1%)	
Postoperative KPS Skore			
90-100	21(42.9%)	26 (54.2%)	
70-80	19 (38.8%)	14 (29.2%)	0.525
<70	9 (18.4%)	8 (16.7%)	
Postoperative KPS Score			
Improved	9 (18.4%)	12 (25%)	

| Stable | 26 (53.1%) | 24 (50%) | 0.745 |
| Deterioration | 14 (28.6%) | 12 (25%) | |

4. Discussion

This study emphasizes the advantages of using SF in combination with IOUS compared with SF alone for glioblastoma resection. Although the SF with IOUS group achieved a higher GTR rate (83.3%) than the SF alone group (67.3%), the difference did not reach statistical significance (p = 0.112). Nevertheless, SF with IOUS demonstrated improved accuracy in identifying residual tumor tissue intraoperatively, as indicated by its stronger agreement with postoperative MRI findings ($\varkappa$: 0.625 vs. 0.447) and NPV of 90.9%. Furthermore, the survival analysis revealed a significant benefit, with the SF with IOUS group showing a longer estimated mean survival (24 months) than the SF group (14 months) (p < 0.001). Postoperative neurological deficits were observed at comparable rates in both groups (22.4% vs. 20.8%, p > 0.05), suggesting that incorporating IOUS does not increase the risk of adverse effects.

Fluorescein-guided surgery is widely used because of its affordability and high intraoperative staining rates of >90% [33–35]. However, its dependence on BBB disruption may reduce its specificity in distinguishing tumor tissue [36,37]. A primary challenge with SF use is that fluorescence can spread into surrounding areas over time, making it harder to delineate tumor margins accurately [38]. Conversely, IOUS provides real-time imaging that is independent of BBB integrity, providing detailed insights into tumor depth and its relation to vital neurovascular structures [39]. This makes IOUS a valuable addition to intraoperative imaging, applicable across various tumor types.

The increasing adoption of IOUS in neurosurgery is particularly beneficial when tumor boundaries are unclear. Unlike fluorescence-based techniques, IOUS allows for continuous intraoperative evaluation of the residual tumor, providing immediate feedback on resection progress [27]. This is particularly advantageous when en bloc resection is not possible, because IOUS facilitates piecemeal removal and ensures maximal safe resection. Previous research has demonstrated that the performance of IOUS is comparable to that of intraoperative MRI in detecting residual tumor in pediatric brain tumors, further supporting its use in surgical planning [39]. Moreover, another advantage of IOUS is that, compared with IOMRI and IOCT, it can be easily used in several centers due to its lower cost, ease of use, and broader accessibility.

Despite its benefits, IOUS has certain drawbacks. Image quality may be affected by artifacts caused by surgical debris, blood accumulation, and tissue distortion, potentially causing misinterpretation [40]. Techniques such as using smaller phased array probes and alternative ultrasound-compatible fluids with acoustic properties more similar to those of brain tissue have been proposed to improve image resolution [41]. Furthermore, IOUS is highly dependent on the operator's experience and skill level, requiring adequate training to optimize its application. Although some institutions involve radiologists for intraoperative interpretation, in several settings, including ours, the evaluation is solely conducted by the operating neurosurgeon. Expanding IOUS training in neurosurgical education may improve its effectiveness and broader clinical application.

Accurately defining tumor margins intraoperatively is vital for maximizing safe resection, which is a major determinant of survival in patients with glioblastoma [42]. Various intraoperative imaging techniques, including neuronavigation, intraoperative MRI, and fluorescence-guided surgery, have been introduced to improve surgical outcomes. Although 5-ALA has been demonstrated to provide high tumor specificity, SF remains a more cost-effective option, despite its lower specificity [31]. Remarkably, previous studies suggest that SF does not significantly influence median survival, emphasizing its role as a surgical aid rather than a determinant of long-term prognosis [31].

This study has some limitations. Computerized volumetric analysis was not performed, which could have provided a more precise quantification of tumor resection. The determination of residual tumor solely relied on contrast enhancement in postoperative MRI, without intraoperative histopathological confirmation. Furthermore, SF and IOUS interpretation was subjective and

depended on the surgeon's expertise. The retrospective nature of the study and the relatively small patient cohort introduce a potential selection bias, which may limit the generalizability of the findings.

5. Conclusions

Combining SF with IOUS may improve intraoperative tumor visualization and resection accuracy without increasing postoperative complications. Although SF facilitates rapid tumor identification based on BBB disruption, IOUS provides real-time depth evaluation and continuous monitoring throughout the surgical procedure. This complementary approach optimizes surgical decision-making and may result in better resection outcomes in patients with glioblastoma. Future studies should aim at refining imaging protocols, minimizing artifacts, and further validating the effectiveness of SF and IOUS in improving GTR rates. As intraoperative imaging continues to evolve, integrating multiple modalities may become a standard practice in neurosurgical oncology to improve patient outcomes.

Abbreviations

IOUS intraoperative ultrasonography
SF sodium fluorescein
GTR gross total resection
IDH-1 isocitrate dehydrogenase-1
MRI magnetic resonance imaging
IOMRI intraoperative magnetic resonance imaging
IOCT intraoperative computed tomography
5-ALA 5-aminolevulinic acid
HGGs high-grade gliomas
PFS progression-free survival
OS overall survival
BBB blood–brain barrier
PPV positive predictive value
NPV negative predictive value
KPS Karnofsky Performance Status
IQR interquartile range
CI confidence interval

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