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Neurosurgical tools to extend tumor resection in hemispheric low-grade gliomas: conventional and contrast enhanced ultrasonography

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Abstract
Purpose Pediatric low-grade gliomas (LGGs) are the most frequent solid tumor in childhood. Based on an increasing number of literature reports, maximal safe resection is recommended as the first line of treatment whenever possible. However, distinguishing tumor tissue from the surrounding normal brain is often challenging with infiltrating neoplasms, even with the assistance of intraoperative, microscopic and conventional neuronavigation systems. Therefore, any technique that enhances the detection and visualization of LGGs intraoperatively is certainly desirable.
Methods In this paper, we reviewed the role of intraoperative conventional ultrasound and contrast-enhanced ultrasound (CEUS) as a tool for extending tumor resection in LGGs. Moreover, our experience with this technology is reported and discussed.
Results Both B-mode and CEUS are helpful in highlighting LGGs, detecting tumor margins and providing additional information such as vascularization, thus improving the safety of a more radical resection.
Conclusions Although the full potentialities of the method are yet to be explored, intraoperative ultrasound is a promising tool in oncologic surgery and LGG surgery.

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Introduction

Pediatric low-grade gliomas are the most frequent solid tumor in childhood which 85 % of these tumors are represented by pilocytic astrocytomas (WHO grade I); while the remaining LGGs which are diffuse, infiltrating astrocytomas and pleomorphic xanthoastrocytomas, diffuse oligodendroglioma. or oligoastrocytoma are rare in children. Surgical treatment of these lesions is often indicated and a maximal safe resection is usually advocated, when feasible, as the most effective initial treatment of these tumors. Adjuvant treatments, such as external beam radiation, chemotherapy, and targeted therapy are considered in cases of progressive or recurrent lesions not amenable of surgical treatment [2, 20].

Indeed, in reviewing the current data regarding the surgical management of low-grade gliomas, there is still lack of class I evidence concerning the need for maximum extent of resection in order to achieve optimal outcome and quality of life for the patient. However, an expanding body of recent literature seems to support that maximal safe resection strongly correlates to longer survival and improved quality of life.

In 2012, Jakola et al. published their data from a population-based cohort study, performing a retrospective analysis of 153 consecutive patients receiving different management at two neighboring hospitals. All the patients were affected by low-grade glioma, and, depending on the hospital that served their region, they received early resection versus a "biopsy and wait-and-see" approach. End point was the overall survival, with a median follow up of 7 years. Overall survival was superior with early resection, with an estimated 5-year

survival of 60 % after biopsy and 74 % after surgery [14].

In a 2014 paper, Hervey-Jumper et al. reviewed data from 21 studies since 1990 where the effect of extent of resection on patient survival and tumor progression for LGG was examined. Their conclusion was that literature shows a mean survival benefit for LGG from 61.1 to 90 months with maximal resections [12].

An important issue affecting the quality of life of patients harboring a LGG is the presence of seizures.

Reports estimate an overall seizure incidence as high as 75–100 % in children, and adults with DNET and low-grade astrocytomas [39]. In fact, in children, the most common pathologic subtypes associated with seizures are low-grade glioma, oligodendroglioma, ganglioglioma, pleomorphic xanthoastrocytoma, and dysembryoplastic neuroepithelial tumor (DNET) [37]. Literature seems to suggest that a greater extent of resection is associated with a better seizure control [7]. In fact, Ullrich et al. reported that subtotal tumor resection was identified as a predictor of seizure recurrence [38].

Given that EoR is crucial for outcome, then precise localization of the tumor and definition of its margins related to the surrounding edema and healthy brain parenchyma are mandatory in order to avoid unnecessary violation of normal brain tissue and minimize the risk of neurological complications for the patient. Surgery deals with living tissues; as such, they are subjected to physiological and mechanical phenomena that constantly change their status. Preoperative imaging has represented (and actually still is) the main tool for surgical planning and surgical intraoperative guidance; in fact, even modern neuronavigation systems mostly rely on preoperative images. However, as a skull bone flap is elevated and surgical resection proceeds, the intracranial content may shift, thus, making any navigation system based on preoperative imaging often unreliable and inaccurate. Therefore, when delineation of the lesion and its margins becomes crucial for improved outcome, any technique that allows for intraoperative, real-time visualization is certainly desirable. With the development of technological advances in neurosurgery, different tools have been recently introduced in an effort to address this issue. Intraoperative CT and intraoperative MRI allow for repeated scans during surgery with the accuracy of an imaging modality that is familiar to the surgeon. Optical imaging, relying on the visualization of fluorophores under specific filters, allows for real time detection of blood vessels and pathological tissues. Intraoperative ultrasound is a relative new tool for neurosurgeons, which is recently becoming more popular since it has the advantage of being a cheap, real-time, reproducible and time-saving technique, with significant potential future applications.

Conventional ultrasound

The B-mode or brightness mode is considered the conventional modality for ultrasonography. Sections of the insonated field are depicted using a gray scale codification. The brightness of the examined tissues' echoes can be evaluated as hyperechogenic, hypoechogenic, and isoechogenic. Cerebral structures have specific echographic features and so have different pathological tissues.

There is still an ongoing debate about the reliability of conventional ultrasound for intraoperative visualization of LGGs. Some authors feel that for LGGs, tumor visualization and borders identification are not optimal by means of standard B-mode US [1, 36].

Conversely, other studies depicted conventional US as a useful intraoperative tool for tumor definition and EoR enhancement [9, 16, 40].

Gerganov et al. [8] compared the image quality of intraoperative MRI and US in a series of 11 patients operated on for LGG removal. They found that tumor location could be accurately assessed in all patients with both methods, and tumor borders could be visualized in almost all cases; however, US accuracy decreased as surgery proceeded [8].

Similar results were more recently reported by *Petridis* et al. [22]; the authors retrospectively evaluated 34 patients operated on for LGG, showing a 0 % failure rate in localizing LGG under ultrasound guidance. However, after tumor removal, they found that the quality of standard ultrasound in identifying tumor remnants in the margins of the cavity decreased significantly and became unreliable.

Along with *Šteňo* [35], we do not confirm this latter finding. In fact, there are many suggested ways to overcome this issue (i.e., MRI preoperative and ultrasound real-time fusion-imaging—inserting a miniature ultrasound probe in the cavity using an artifact-reducing acoustic coupling fluid). In our experience, we also found that insonating the surgical cavity orientating the probe so that the ultrasound beam crosses the surrounding parenchyma, instead of passing throughout the very cavity, may help in reducing the artifacts from the blood and debris.

Moreover, the probes utilized by the authors above were *Convex* type probes. *Coburger* et al. [4] conducted a prospective non-randomized study in order to assess the accuracy of *Linear* array ultrasound in comparison to conventional intraoperative ultrasound and intraoperative MRI [4]. In their study, they enrolled 13 patients harboring a WHO grade II glioma; after removal, they checked the cavity for residual tumor with iMRI, conventional US (cioUS), and linear array ultrasound (lioUS); furthermore, they performed a total of 30 biopsies from the resection cavities and correlated the histopathological findings with the intraoperative images. Their results showed that iMRI had the highest sensitivity in detecting residual tumor (83 %), and similarly the sensitivity of

lioUS was 79 % while cioUS showed only a 21 % sensitivity. In other words, they found that images from iMRI and from lioUS strongly correlated, and both were significantly linked to final histopathology. This correlation between MRI and lioUS is an important finding, since different previously published studies showed higher resection rates in LGG surgery when the use of iMRI was involved [10, 17, 21, 34].

Contrast-enhanced ultrasound (CEUS)

Ultrasound Contrast Agents (UCAs) are purely intravascular contrast agents, thus, differing from MRI contrast agents, which can diffuse in the interstitial space. They are made of a microbubble (MB) structure (gas stabilized by a shell) and can add different biological information to standard B-mode, bearing the capability to visualize blood flow and vasculature tree in a structure/organ. Second generation UCAs permit a continuous study of structure/organ for several minutes, dynamically evaluating in real-time the enhancement patterns.

Over the years, a huge number of studies have described the UCA application in the liver and many other organs, and recently, literature regarding the use of UCAs in neurosurgery is growing [6, 11, 13, 15].

However, there is still little literature addressing the utility of CEUS in brain tumors and specifically for LGG.

Our group has previously published numerous studies regarding the intraoperative use of CEUS for different neurosurgical pathologies. In particular, two of them were focused on the characterization of brain tumors, by means of standard B-mode ultrasound and CEUS.

In one paper [31], we analyzed 71 patients harboring different intracranial lesions, 16 of which were low-grade gliomas. We performed an unprecedented intraoperative qualitative analysis comparing CEUS with B-mode US imaging and preoperative MRIs and correlating data to histopathology. We used a 3-to 11-MHz linear US navigated probe. We initially observed the lesions on B-mode imaging, defining them as highly hyperechoic, mildly hyperechoic, or isohypoechoic compared with the surrounding normal brain parenchyma. All LGGs appeared mildly hyperechoic compared with brain parenchyma. We observed that in B-mode, the brain/tumor interface was not always clearly visible everywhere and was indistinguishable from edematous brain parenchyma.

Tumor CE was then evaluated in an offline setting following the EFSUMB criteria on CEUS [23] performing an inter-observer, semi-quantitative analysis considering several parameters: timing (arterial and venous phase), degree of Contrast-Enhancement (CE) comparison with brain parenchyma, and contrast distribution (centripetal/centrifugal pattern, visibility of afferent/efferent vessels, intralesion vessels, cystic/ necrotic areas).

Surprisingly, we found a subtle but clearly visible CE also in low-grade gliomas, which usually lack CE on MRI. In low-grade gliomas, CEUS showed scattered and dotted CE with slow vascular phases in all cases, even if borders were not always clearly distinguishable from healthy brain tissue.

In another paper [30], we focused on a series of patients affected by glioma. Among the 69 patients analyzed, 22 harbored a LGG (18 astrocytomas and 4 oligodendrogliomas). Semiquantitative analysis confirmed our previous results regarding vascular phases and timings after contrast injection.

The overall picture shows that in B-mode, the main differences between lesions at different grades of malignancy are the degree of hyperechogenicity when compared to the surrounding parenchyma, the presence of cystic/necrotic areas, and a more or less defined brain/tumor interface. On CEUS, lower grades were characterized by gradually less intense CE and less defined tumor borders when compared to higher grades. Nonetheless, a slight but well-defined CE was observed in low grades too, where preoperative MRI did not show any enhancement. Moreover, CEUS provided information about intra/perilesional vessels.

We also reported that in more than one case, preoperative MRI oriented towards the diagnosis of LGG, showing a lesion hyperintense in FLAIR, hypointense in T1-weighted sequences, with no contrast uptake at all. During tumor removal, CEUS analysis showed a few spots of contrast uptake; the final hystopathological diagnosis was grade III astrocytoma, thus, suggesting that CEUS might also have higher accuracy in correlating with histology. This could represent a very important feature, since CEUS may guide the surgeon in deciding which area may be more representative, for example in case of biopsy.

More recently, *Cheng* et al. [3] performed conventional ultrasound and CEUS in a series of 88 patients, 38 of them harboring a low-grade glioma. They investigated the relationship between CEUS parameters in different grades and MicroVascular Density (MVD) and Vascular Endothelial Growth Factor (VEGF) in the corresponding pathological samples. Firstly, they found that the enhancement at CEUS of LGGs was higher than the normal brain and surrounding edema. More precisely, the Absolute Peak Intensity (API) of LGGs was significantly higher than the normal brain and surrounding edema. Moreover, API of glioma showed a positive correlation with MVD.

Given these preliminary results, CEUS appears a promising tool in visualizing and defining LGGs, along

with the other intra-operative US modalities. However, there is still little literature addressing the role of CEUS in low grade-gliomas, especially in pediatric populations. If on one side, this is due to the relatively recent introduction of CEUS in neurosurgery. On the other side, this is undoubtedly ascribable to the fact that at present, there is no UCA approved in Europe for intravenous use in children while it is now considered a safe drug for adult use. Therefore, any utilization in pediatric population comes in an off-label setting, and should be tailored case by case.

Few studies addressed the problem of safety of ultrasound contrast agents in younger patients [5, 18, 19, 24, 32, 33].

In 2012, *Riccabona* [32] reported the results from a large European questionnaire-based survey investigating the safety of the intravesical and IV use of SonoVue (sulfur hexafluoride, Bracco) ultrasound contrast agent in children. Of the 5079 examinations in children (age mean: 2.9 years; range: birth–18 years), 948 utilized intravenous application. The data recorded overall depict a high safety profile of the UCA SonoVue®, which is the most widely used UCA in Europe.

Coleman et al. [5] in 2014 published their data about intravenous administration of UCA in pediatric population. They performed a total of 134 CEUS examinations in 34 children and young adults affected by malignancies. Their conclusion is that IV-administered ultrasound contrast agents have an excellent safety profile in children.

In 2016, *Rosado and Riccabona* [33] analyzed the existing literature regarding the off-label use of UCAs for intravenous applications in children for a total of 540 reported cases; their finding support the conclusion that the IV use of US contrast agents in children is safe and feasible.

Therefore, we believe together with the radiological community, that there is a need for the use of CEUS in pediatric population to be approved.

Conclusions

Intra-operative ultrasound (ioUS) has been used in neurosurgery for many years. Recent technological advances have refined ultrasound spatial resolution and added new useful features for better tissue and vessel delineation. All these advances have fostered the use of ioUS, which is now becoming more widespread in the surgical community, both as a stand-alone tool or in a synergistic use with other imaging modalities. Our group introduced intraoperative ultrasound in the routine practice since 2007, and since then, we feel that it has become an essential tool in intraparenchymal tumor-surgery. Technical improvements and increasing confidence with the technique are leading to overcome limitations and extend the indications to the use of this image modality.

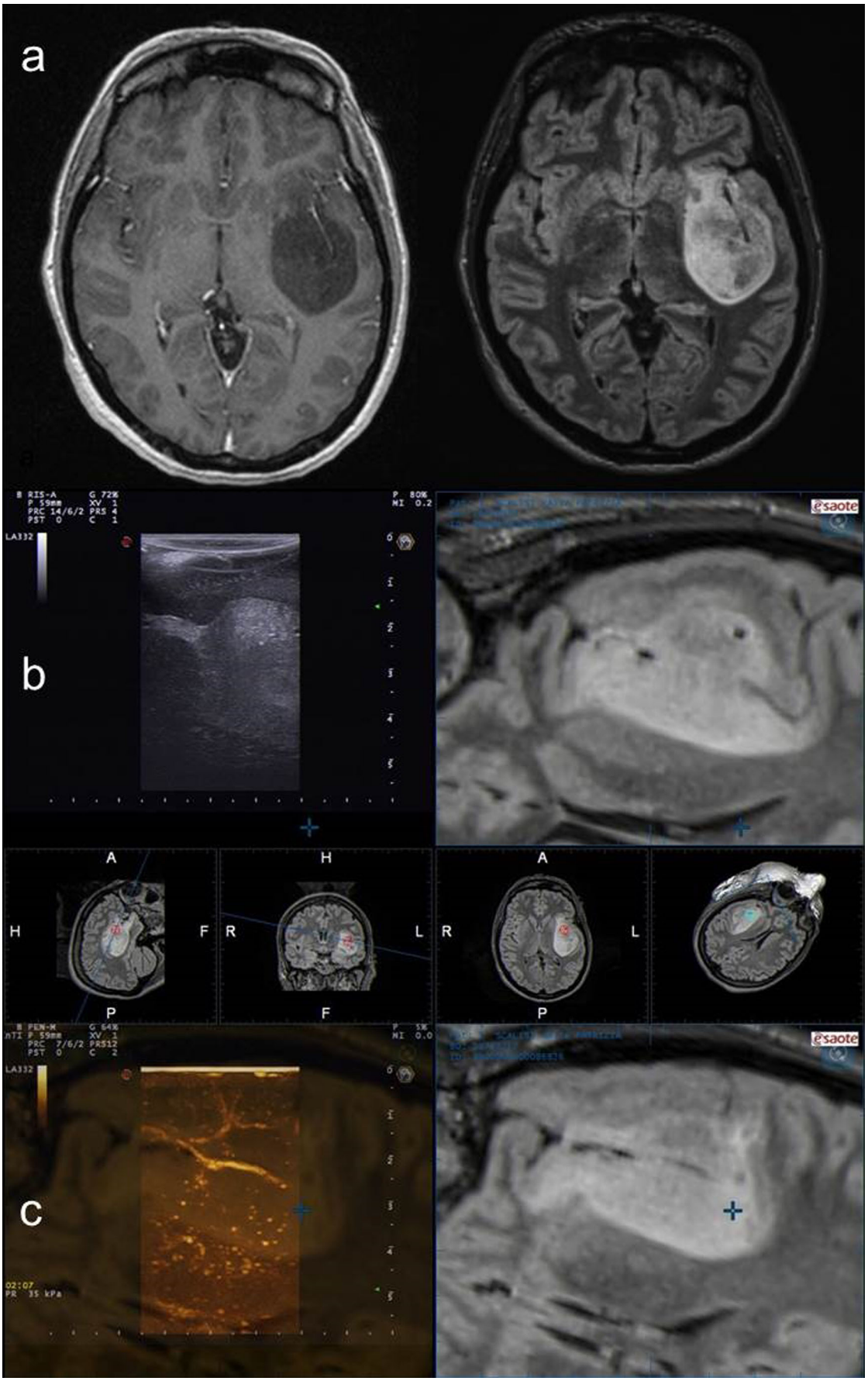
Relying upon our experience with low- grade gliomas, we feel that either standard B-mode and CEUS could be a useful tool for extending safely tumor resection. B-mode helps in detecting the tumor at the beginning of the surgical removal, orientating the surgeon when the cortical plane is still intact, and guiding the procedure as the resection proceeds. CEUS not only enhances B-mode findings, but also adds some valuable information, for example providing a better definition of tumor borders when peritumoral edema makes them hardly detectable in B-mode. Moreover, it provides unique information about the perfusion of healthy and tumoral tissues, allowing direct visualization of intra- and perilesional vessels, thus orienting the surgical strategy [25, 29].

Finally, neuronavigation along with fusion imaging provides the possibility to superimpose functional MRI and DTI imaging to the real-time US imaging, repeatedly adjusting and re-orienting the navigation system relying on intraoperative images. This allows to proceed with the resection keeping all the available functional information consistent with the actual intraoperative situation, thus, supporting the information from neurophysiological monitoring, and overall improving the safety and the extent of the resection. (Figs. 1 and 2 show two illustrative cases and some features of intraoperative US in LGG surgery).

Of course, ultrasound bears some intrinsic drawbacks.

Firstly, it is an operator-dependent methodic, and as such it could be biased by subjective interpretation. More importantly, the iconography provided, is almost unknown to the neurosurgical community. Neurosurgical training usually leads the surgeon to become familiar with more traditional imaging, such as CT and MRI. Interpretation of US imaging requires a specific training, and cannot be improvised. We found that coupling intraoperative US imaging with preoperative MRI by means of fusion imaging navigation systems, can effectively speed up the learning process, providing a steeper learning curve. In fact this process allows the neurosurgeon to constantly compare the anatomical structure identified at US with the corresponding imaging as shown at MRI [26, 27].

Fig. 1 **a** Contrast-enhanced T1 and FLAIR-weighted axial MRI scans showing a left fronto-temporal-insular low-grade glioma. **b** The US probe is navigated and B-mode scans are displayed in split screen along with the matching preoperative MRI scans. In B-mode the lesion appears brighter (hyperechogenic) compared to the surrounding parenchyma, tumor borders appear well defined. **c** CEUS highlights intralesional vessels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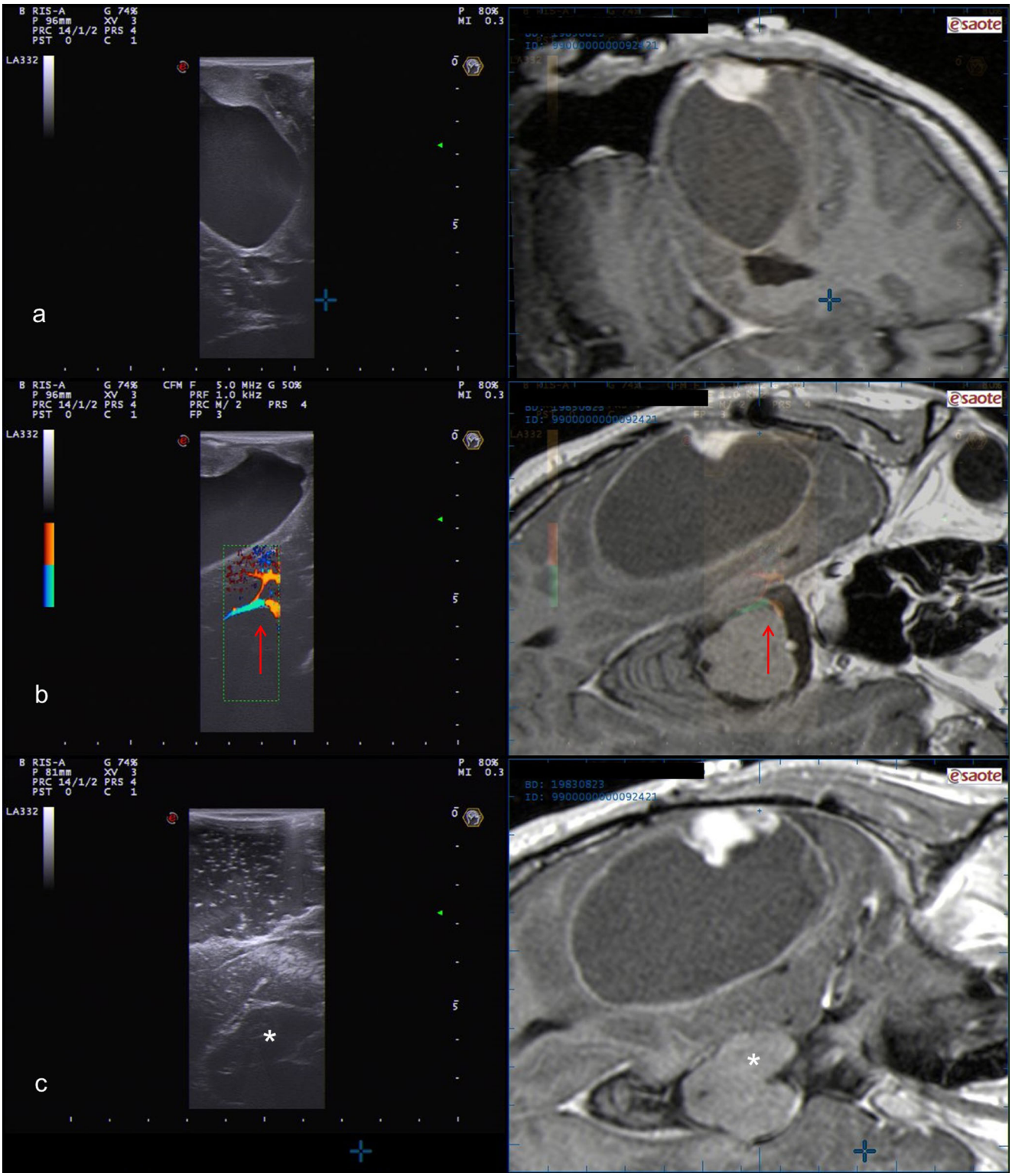

Fig. 2 Intraoperative split-screen displaying a left temporal pilocytic astrocytoma as seen in standard B-mode and in MRI imaging. **a** In B-mode, the solid component appears hyperechogenic comparing to surrounding parenchyma, while the cystic component appears clearly hypoechogenic. **b** Doppler US identifying the left posterior cerebral artery (*red arrows*). **c** At the end of surgical removal the field is filled with saline solution and then insonated, showing no residual tumor. Brainstem is identified as a hypoechogenic structure comparing to surrounding tissue (*white asterisks*)

In conclusion, US is a time-saving, cost-saving, reproducible, real-time imaging technique. It is beyond the purpose of this dissertation to list the several features that are progressively implementing this methodic (i.e., Navigated ultrasound, Doppler ultrasound, Elastosonography) [28]. Surely, the full potentialities of the methodic are yet to be explored, but we feel that intraoperative US is a promising tool in oncologic surgery and in LGG surgery.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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