

## Supplementary Data 1. Technical details and workflow

### The Setup

Since 2018, we have used a commercially available high-end ultrasound scanner bk5000 (BK Medical, Harlev, Denmark) digitally integrated with Brainlab KICK navigation with ultrasound navigation software (Brainlab AG, Munich, Germany) to perform navigated three-dimensional ultrasonography (n3DUS). Fig. 1 shows the layout of the operating room with the equipment setup during a surgery. The navigation system consists of a central navigation computing unit consisting of a navigation planning interface and an optical infra-red detection camera which detects the reflecting spheres affixed to the reference frame and the navigation pointer along with ultrasonography (US) probe using a passive optical reflection detection technique for localization. The US system (covered with a sterile drape) is positioned adjacent to the navigation system and within reach of the operating surgeon for direct access of the controls. The US probes selected for the navigation are sterilizable but can also be covered with a sterile drape to ensure optimal acoustic coupling using sterile gel. Note that all probes may not be navigable and different systems may offer different probes for this purpose. We generally use a curved linear probe with a small footprint (29×10 mm) and frequency range of 5–13 MHz (Craniotomy N13C5 transducer, BK Medical) which is also used for navigated ultrasonography. The transducer parameters are as follows:

Footprint: 29×10 mm

Acoustic aperture: 29×6 mm

No. of elements: 160

Radius of curvature: 25 mm

Transverse focal length (typical): 35 mm

Image plane focal length: variable

Focal range (depth): 10–80 mm

For intracranial use, the highest frequency band is selected (usually this is an auto-select feature) with a frequency of 8 MHz used for most subcortical tumor surgeries (higher range for more superficial structures and vice versa). The depth of the US scan is generally

kept at 6–7 cm (but may vary depending on the location of the tumor from the surface) which is sufficient to encompass the tumor and adequate surrounding landmarks. Focus is set at the centre of the tumor to ensure optimal resolution of image at this depth. A customized tracking device can be snugly affixed to the transducer allowing it to be detected by the navigation system (Fig. 2). Digitally integrated US and navigation system allows accurate one-time calibration of the US probe during installation of the system. This integrated system now allows display of the real-time two-dimensional ultrasonography (2DUS) image on the navigation screen along with the co-display of the corresponding magnetic resonance imaging plane besides providing for the option of acquiring a n3DUS as described below (Figs. 4–8).

### Workflow

The patient is positioned in operating theatre with the head fixed rigidly in head-clamps such that the post-resection cavity is as vertical as possible in order to be filled optimally with saline to allow a good post-resection US scan at the end of the surgery (Fig. 3). The navigation reference frame is attached to the head-holder rigidly in a routine fashion. The navigation system is pre-loaded with the volumetric T2 and T1 sequences of magnetic resonance for marking region of interest and B0 diffusion weighted sequences for fibre tracking. The spatial relationship between the two different co-ordinate systems of magnetic resonance imaging and patient in 3D space is then created by registering the patient using surface matching system for planning craniotomy following which craniotomy is performed in standard fashion. The US system is connected to the navigation digitally. When using 2DUS, the live image is seen on the navigation screen (identical to the US machine screen). Once the tracker is affixed onto the probe, it becomes navigable. Both navigated two-dimensional ultrasonography and n3DUS can then be performed as outlined in the paper. Each time the US is acquired, the resection cavity is filled with adequate saline to ensure acoustic coupling. There is no fixed amount of saline required. This depends on the size of the cavity.