ABSTRACT: Background: The brain functions as an integrated multi-networked organ. Complex neurocognitive functions are not attributed to a single brain area but depend on the dynamic interactions of distributed brain areas operating in large-scale networks. This is especially important in the field of neurosurgery where intervention within a spatially localized area may indirectly lead to unwanted effects on distant areas. As part of a preliminary integrated work on functional connectivity, we present our initial work on diffusion tensor imaging tractography to produce in vivo white matter tracts dissection. Methods: Diffusion weighted data and high-resolution T1-weighted images were acquired from a healthy right-handed volunteer (25 years old) on a whole-body 3 T scanner. Two approaches were used to dissect the tractography results: 1) a standard region of interest technique and 2) the use of Brodmann’s area as seeding points, which represents an innovation in terms of seeds initiation. Results: Results are presented as tri-dimensional tractography images. The uncinate fasciculus, the inferior longitudinal fasciculus, the inferior fronto-occipital fasciculus, the corticospinal tract, the corpus callosum, the cingulum, and the optic radiations where studied by conventional seeding approach, while Broca’s and Wernicke’s areas, the primary motor as well as the primary visual cortices were used as seeding areas in the second approach. Conclusions: We report state-of-the-art tractography results of some of the major white matter bundles in a normal subject using DTI. Moreover, we used Brodmann’s area as seeding areas for fiber tracts to study the connectivity of known major functional cortical areas.

RÉSUMÉ: La tractographie dans l’étude du cerveau humain : perspective neurochirurgicale. Contexte : Le cerveau fonctionne comme un organe constitué en multiréseaux intégrés et les fonctions neurocognitives complexes ne sont pas restreintes à une seule zone du cerveau. Elles dépendent d’interactions dynamiques de différentes régions du cerveau opérant sur des réseaux de grande envergure. Ceci est particulièrement important dans le domaine de la neurochirurgie où une intervention à l’intérieur d’une zone très localisée peut provoquer indirectement des effets indésirables à distance. Nous présentons, dans le cadre d’un travail intégré préliminaire sur la connectivité fonctionnelle, nos travaux initiaux sur la tractographie par IRM de diffusion pour obtenir in vivo la dissection de faisceaux de la substance blanche. Méthode : Des données de l’IRM pondérée en diffusion et des images de haute résolution pondérées en T1 ont été acquises chez un volontaire sain droitier de 25 ans au moyen d’un scanner T3 du corps entier. Deux approches ont été utilisées pour disséquer les résultats de la tractographie : 1) une technique standard ciblant une région spécifique et 2) l’utilisation de la zone de Brodmann comme point d’essaimage, ce qui constitue une innovation. Résultats : Nous présentons des images de tractographie tridimensionnelles. Le faisceau unciné, le faisceau longitudinal inférieur, le faisceau fronto-occipital inférieur, le faisceau pyramidal, le corps calleux, le cingulum et les radiations optiques de Gratiolet ont été étudiés par la méthode d’essaimage conventionnelle alors que les zones de Broca et de Wernicke ainsi que les cortex primaires moteurs et visuels ont été utilisés comme zones d’essaimage dans la deuxième approche. Conclusions : Nous rapportons des résultats de tractographie par IRM de diffusion, une technologie de pointe, de certains des faisceaux importants de la substance blanche chez un sujet normal. De plus, nous avons utilisé la zone de Brodmann comme zone d’essaimage afin d’étudier la connectivité des zones corticales fonctionnelles majeures connues.

And what are the repercussions of a surgery to remove such a tumour? With regards to these considerations, brain surgery is still in its infancy; much progress remains to be accomplished to fully optimize the reach of the surgical gesture so that tumour resection is maximized, whereas it remains within respect of the unique neurophysiological limits of a patient’s functional anatomy. Given that function in one brain area highly depends on the function of distant areas (connectivity), surgical intervention not only affects the area surrounding the tumor, but also distant areas which are coupled to it. 

Although initial knowledge acquisition of brain functionality allowed identification of specified punctual anatomical cortical areas such the primary motor cortex, the primary sensory and visual cortex, Wernicke’s and Broca’s areas, as well as the identification of the major white matter tract bundles (e.g. cortico-spinal, arcuate fasciculus, corpus callosum), it is increasingly recognized that the brain functions as an integrated multi-networked organ. Thus, complex neurocognitive functions are most likely not attributed to a single brain area but rather depend on the dynamic interactions of distributed brain areas operating in large-scale networks.

Interestingly, as these networks are the results of countless connections from different areas of the brain and as these connections are genetically as well as environmentally determined and are in a constant state of flux, they thereby present a certain degree of variability from patient to patient. If we add the remodeling induced by a growing glioma as a factor, then it becomes obvious that the functional anatomical situation cannot be easily ascertained, other than by an individual in-depth study for each patient.

Recently, the conjunction of different tools has allowed a better in vivo study of the structure of the human brain. One such tool is diffusion Magnetic Resonance Imaging (dMRI), allowing the study of white matter tracts, using a paradigm known as diffusion tensor imaging (DTI) and tractography. This approach uses the inherent diffusion properties of water molecules, which, in unrestricted structures, follows a Brownian motion; that is, a random movement based on high speed collisions. The summation of this diffusion process should be equal in all directions, thereby producing isotropic diffusion. Whenever this diffusion process is unequal and overall results in collisions, the summation of this diffusion process should be equal in all directions, thereby producing isotropic diffusion. This approach uses the inherent diffusion properties of water molecules, which, in unrestricted structures, follows a Brownian motion; that is, a random movement based on high speed collisions. The summation of this diffusion process should be equal in all directions, thereby producing isotropic diffusion. If we add the remodeling induced by a growing glioma as a factor, then it becomes obvious that the functional anatomical situation cannot be easily ascertained, other than by an individual in-depth study for each patient.

The brain was skull stripped from the T1-anatomy, which was aligned with the Talairach stereotactical coordinate system. The 21 images without diffusion weighting were placed at the beginning of the sequence and after each block of 10 diffusion weighted images to provide anatomical reference for offline motion correction. The measurement of 72 slices with a thickness of 1.7 mm covered the whole brain. Random variations in the data were reduced by averaging the data from different integrated systems of the brain, we fuse tractography results with Brodmann areas. We recently undertook an ambitious study integrating dMRI and functional MRI (fMRI) images as well as neuro-cognitive studies of patients operated on for malignant glial tumours. We hereby present our initial work on dMRI and DTI. In this work, we first illustrate large fiber bundles that are effectively recovered with DTI tractography. Then, in an effort to explore different integrated systems of the brain, we fuse tractography results with Brodmann areas. In this setting, the Brodmann areas have been used as regions of interest to characterize different fiber tracts. This study was performed in a normal brain, and it is being used as a basis to transfer this concept to the individual analysis of each brain tumour patient we treat.

**DIFFUSION MRI data acquisition & tractography**

Diffusion weighted data and high-resolution T1-weighted images were acquired from a healthy right-handed volunteer (25 years old) on a whole-body 3 T Magnetom Trio scanner (Siemens, Erlangen) equipped with an 8-channel head array coil. The spin-echo echo-planar-imaging sequence (TE = 100 ms, TR = 12 s, 128x128 image matrix, FOV = 220x220 mm²) consists of 60 diffusion encoding gradients with a b-value of 1000 s/mm². Seven images without any diffusion weightings were placed at the beginning of the sequence and after each block of 10 diffusion weighted images to provide anatomical reference for offline motion correction. The measurement of 72 slices with a thickness of 1.7 mm covered the whole brain. Random variations in the data were reduced by averaging the data from three acquisitions, resulting in an acquisition time of about 45 minutes (min). The signal-to-noise ratio (SNR) in the white matter of the images averaged from three acquisitions was estimated to be approximately 24.

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Diffusion tensor imaging and tractography are fraught with certain pitfalls that can severely bias the results. This technique is still emerging, and new acquisition schemes such as high angular resolution diffusion imaging (HARDI) and analysis algorithms are still being developed to circumvent those pitfalls.
Tractography dissection

Two different approaches were used to dissect the tractography results. For the classical white fiber bundles, a standard region of interest technique was used\(^9\,20\). In this approach, selection boxes are placed along the expected trajectory of known white matter tracts using anatomical atlases as general guides\(^21\). Typically, a master selection box allows the gross identification of the white matter bundle of interest, whereas a second exclusion box allows the elimination of aberrant white fibers, not belonging to the studied fasciculus. This “two-regions of interest” approach has initially been described in\(^22\), and has been detailed in a seminal paper by Catani et al\(^9\). Obviously, the placement of these ‘region of interest boxes’ is an iterative and dynamic process. For further characterization, the FiberNavigator (open source software: http://code.google.com/p/fibernavigator/)\(^19\) also allows the inclusion of other selection boxes that can be used with AND/NOT logical operators, thereby increasing the power of the fiber dissection. This methodology was used for all but two tracts: the superior longitudinal fasciculus (SLF) and the corpus callosum. Because the SLF branches to so many areas, the inclusion of a second region of interest (ROI) box significantly affected the density of identified fibers, and conducted to the exclusion of fibers actually belonging to this bundle. Thus, only one master ROI box was used for the SLF. Given the nature of the corpus callosum, multiple master boxes were used in the genu, body and splenium of the corpus callosum to allow a specific selection of these fibers, excluding nearby unrelated fibers.

The second approach that was used represents an innovation in terms of fiber selection strategy, and was used to test the limit and potential of DTI tractography. We used Brodmann’s area as seeding points to study the connectivity of known major functional cortical areas. To do so, the 48 Brodmann areas derived from MRICro (publicly available neuroimaging software)\(^23\) were warped into the T1 space of our dataset using rigid registration (using FSL). At the same time, a white matter mask was created with FSL. This mask was finally intersected with the warped Brodmann areas, and only the areas intersecting the white matter were kept. These remaining Brodmann areas were then integrated in the FiberNavigator for fiber selection as Volume of Interests (VOIs). Thus, each Brodmann area could be used as ROI for tractography dissection. Interestingly, this approach was fruitful, and allowed the study of white matter bundles emerging from Brodmann’s areas and in particular, from the motor, sensory and visual systems, as well as Broca and Wernicke’s areas.

RESULTS

Results are presented as illustrations of the tract obtained, and a brief description of current knowledge on the function of these tracts. We will first look at association fiber systems, followed by commissural and projection systems. Finally, results obtained by using the Brodmann areas as seeding points will be presented.

Superior longitudinal fasciculus (SLF) or arcuate

The SLF is part of the longitudinal association fiber system, which lays connections between the frontal lobe and other areas of the ipsilateral hemisphere (Figure 1)\(^24\). The uncinate, and fronto-occipital fasciculi are other components of this longitudinal association fiber system. The SLF fiber bundle connects perisylvian areas in the hemisphere (frontal, temporal and parietal). As summarized by Catani et al\(^9\), this bundle system can be divided in two components: 1- longer fibers located medially in the fasciculus which mostly connects the lateral frontal cortex with dorsolateral and temporal cortex, and 2- shorter U-shaped fibers running more laterally and mostly connecting fronto-parietal, parieto-occipital and parieto-temporal cortex. The SLF is considered to be the largest fiber bundle system in the brain. It serves a function in higher motor control, whereas its role in language is not firmly established when considering the dominant hemisphere in the literature\(^9,24,25\). Our findings using the Brodmann areas seeding from Broca do reproduce a white matter tract that closely resemble the SLF, thus suggesting that this bundle could indeed participate in language functionality in the dominant hemisphere. Interestingly, it is clearly involved in visuo-spatial processing when considering the non-dominant hemisphere\(^26\).

Uncinate fasciculus

This fasciculus connects the anterior region of the temporal lobe with the orbito and polar frontal cortices (Figure 2). The precise cortical connections in the temporal lobe remains undetermined, with connections to the temporal pole, the amygdala, the hippocampal formation and the superior and
middle temporal gyri all described\textsuperscript{27-29}. A recent study has suggested that naming famous people could be the most relevant task in which this bundle is involved, when considering the dominant hemisphere\textsuperscript{30}. Papagno et al supported this conclusion\textsuperscript{31}. These authors concluded that the uncinate fasciculus was part of a circuitry involved in the retrieval of word form for proper names. The dominant uncinate fasciculus connects the orbitofrontal cortex, involved in face encoding and in processing famous names, to the temporal pole, which is crucial in naming people.

**Inferior longitudinal fasciculus (ILF)**

Using white matter tract dissection, Dejerine demonstrated in 1895 that this bundle connects the temporal with the occipital lobe\textsuperscript{32,33}. Thereby, fibers from the superior, middle, and inferior temporal gyri, as well as from the fusiform gyrus project to the lingula, cuneus, lateral and polar surfaces of the occipital cortex (Figure 3). This tract lines the lateral wall of the occipital as well as of the temporal horns of the ventricular system\textsuperscript{33}. While the function of this fiber bundle remains unclear, recent evidence suggest its potential involvement in object naming, when considering the dominant hemisphere\textsuperscript{34}.

**Inferior fronto-occipital fasciculus (IFO)**

This pathway connects the infero-lateral and the dorso-lateral frontal cortex with the posterior temporal cortex and the occipital lobe (Figure 4)\textsuperscript{35}. Mostly arising in pre-frontal areas, the fasciculus runs immediately superior to the uncinate fasciculus in the frontal lobe and significantly narrows as it travels through the anterior floor of the external capsule, gradually fanning out to the middle and inferior temporal gyri, the lingual and fusiform gyri in the temporal lobe, and the inferior occipital cortex\textsuperscript{9,36}.

Recently, its function has been mapped to the facial recognition of emotions\textsuperscript{37}.

**Cingulum**

The cingulum (Figure 5), which lies just inside of the cingulate gyrus, below the short u-fibers, is a longitudinal
fasciculus running just above the corpus callosum which connects the subcallosal and paraterminal gyri of the frontal lobe with the paracentral lobule, the precuneus and the hippocampus. The cingulum is a component of the limbic lobe.

Corpus callosum

The corpus callosum comprises fibers originating from all areas of the cortex that converge toward the midline and continue to the contralateral hemisphere (Figure 6). While doing so, these fibers form the roof of the lateral ventricle. Functionally, the corpus callosum is divided in three distinct segments: the anterior segment (rostrum and genu), the middle segment (body) and the posterior segment (splenium). The anterior segment connects pre-frontal cortices via the minor forceps, an arcing-shaped bundle containing the radiating fibers. The body of the corpus callosum connects the pre-motor and motor cortices, as well as the parietal and temporal lobes. Fibers crossing through the splenium form the major forceps, while small groups of these fibers travel inferiorly over the inferior horn of the lateral ventricles to connect both temporal lobes. Fibers from the forceps major interconnect the parieto-occipital and calarine regions.

Corticospinal tract (CST)

The major projection fiber tracts from the motor system is probably the most studied tract using DTI. Investigators are looking at the predictive value of characterizing this bundle prior to surgery, as well as after a pathological process such as a stroke. As can be appreciated from Figure 7, one of the weaknesses in adequately defining this bundle pertains to the fibers located laterally on the motor strip of the homunculus. Because of the difficulty in reconstructing the dominant tensor direction in areas where voxels contain numerous fibers crossing, as well as in cases where dominant direction curves abruptly, existing algorithms have difficulty revealing lateral fibers, as they cross the centrum semiovale (3-way fiber crossing through lateral projections of the CC, the SLF and projections of the CST). Therefore, fibers emerging from sensori-motor cortices representing the face are predominantly missing. This explains the fact that this bundle as pictured in Figure 7 presents only its parasagittal fibers when seen in a coronal plane. It is, however, interesting to appreciate those cortico-pontine fibers that transit in the pontine nuclei to gain access via the middle cerebellar peduncle to the dentate nuclei in the cerebellum.

Optic radiation

The visual system is notoriously difficult to study with DTI. In Figure 8, we present the images obtained while studying the optic radiations. As described by Kitajima et al, optic radiations...
have a very low fiber density and wide translucent spaces, thereby greatly impacting the quality of DTI data. Typically, the visualization of the Meyer’s loop and its anterior extension is particularly problematic. However, Figure 8 eloquently illustrates the Meyer’s loop, even if, as discussed by Hofer et al., it is impossible on these images to specifically validate the anterior extension of the loop.

**Brodmann areas as selection regions**

Brodmann’s atlas was published in 1909, and represents the first comprehensive study aiming at defining focal differential areas in the cortex based on the cytoarchitecture, that is the layering of the cells in the cortex of the human brain, as well as myeloarchitecture, or the consequent organization of the subjacent white matter. Using discrepancies in cytoarchitecture and myeloarchitecture, Brodmann identified and defined 44
different areas in the brain. This classification, based on a single human brain represents the first attempt at brain parcellation, and despite its age, these observations have remained valid over time; numerous physiological studies have confirmed the correspondence between cytoarchitectural Brodmann parcellation, and function for several of these areas. Nonetheless, refinements of this parcellation have been proposed, as Brodmann’s approach is not without fault, and distinct functions have been described arising within a single well-defined Brodmann area, such as supplementary motor area (SMA) and pre-SMA within area 6\textsuperscript{a}. However, as this approach still remains the best parcellation technique to this day, we elected to fuse the Brodmann areas into the anatomical T1 space of the subject so that we could then use some of the classic Brodmann areas as selection regions (volume of interest) for fiber tracts overlayed in the FiberNavigator.

For the following study, we used Broca’s and Wernicke’s areas, the primary motor cortex and motor cortices, as well as the visual cortex (Brodmann’s areas 17) as selection regions. Results can be appreciated in Figures 9 to 12.

Selecting from Broca’s area depicts an intricate network of white matter fibers connecting peri-sylvian areas that closely resemble the arcuate fasciculus (Figure 9). As mentioned earlier, we feel that this finding should be in support of a clear role in language function for the arcuate fasciculus (or SLF)\textsuperscript{5}. Selecting from Wernicke’s area produced a more limited network of fibers that ran mostly toward the parietal and occipital lobes and inferiorly toward the temporal lobe, thereby emphasizing connections in and around the parieto-temporo-occipital association cortices (Figure 10).

Evaluation of the motor cortex was performed using prefrontal, primary motor as well as primary sensory cortices. This is in agreement with the concept of a central lobe, regrouping the pre and postcentral gyri as proposed by Yasargil\textsuperscript{49}. This new denomination is based on the fact that both gyri compose a single morphological and functional unit. Thereby, using areas 2, 3, 4, 6, 9, 10 and 11 as selection regions, we could reproduce fiber bundles compatible with the corticospinal tract, displaying a nice fan-like corona radiata, converging toward the internal capsule and also depicting fibers crossing to the middle cerebellar peduncle toward dentate nuclei (Figure 11). The evaluation of the visual system was initially performed by selecting from area 17, the primary visual cortex (Figure 12). Interestingly, the addition of area 18 and 19 as seedings did not significantly add new fibers, implying that area 17 is a central hub for the visual function.

**DISCUSSION**

Brain surgery, in the context of a glial tumour, should still be considered in its infancy. The only accepted principle driving the surgical effort is the maximized surgical resection, while preserving ‘neurological function’\textsuperscript{50-53}. Interestingly, when reviewing studies dedicated to the outcome of glioma surgery patients, one realizes that the clinical surrogates surveyed refer to the Karnovsky score (KPS), the evaluation of speech, motor and sensory function and vision\textsuperscript{50-52}. But what of integrated cognitive functions and more complex tasks? Thanks to the expanding field of brain “hodotopy”, as well as the realization that plasticity is an ongoing active process, our view of the brain has changed from that of pre-determined functional areas that reside within specific confines, to that of a highly integrated

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**Figure 10:** Result obtained when using Wernicke’s area (Brodmann’s area 39 and 40) as the region of interest (red surface) to seed, involving connections between the temporal, parietal and occipital areas.

**Figure 11:** Using the motor system as the region of interest. Primary somatosensory cortex (green), primary motor cortex (red), premotor cortex (yellow), prefrontal cortex (light blue) and orbitofrontal cortex (dark blue) were collectively used as seeding areas. Notice how nicely the cortico-pontine fibers travelling via the middle cerebellar peduncle toward the dentate nucleus are shown.
Ever since the publication of important functional studies such as the brain maps acquired by Ojemann et al in 117 epilepsy patients and showing a great variability in the speech localization, a greater emphasis has been placed on the study of the function, thereby realizing that reliance on topographical anatomy alone is inadequate. Moreover, studies in neuro-oncology are now included as an integral facet of the evaluation of patients quality of life data as well as neuro-cognitive evaluations, leaning toward a more inclusive and integrated view of the patient as an individual-whole.

The concept of brain functioning now involves a dynamic organization of networks connecting several cortical and subcortical areas of the brain. Only through detailed physiologic and functional characterization will we be able to adequately define the extent of these functional networks. This endeavour should eventually be performed for each patient considered for surgery, as the situation is likely to be unique in each patient, especially those in which an extensive remodelling of white matter fibers related to the presence of a glial tumour took place. It is remarkable that even now, patients are operated on with a complete disinterest and incapacity in formally characterizing the impact of a surgery on complex neurocognitive tasks domains, such as executive functions. Different technologies can assist in this task, be it the use of per-op cortical and subcortical stimulation, functional MRI and diffusion MRI. Likely, a combination of these approaches, combined with formal neuro-cognitive testing will eventually lead to the acquisition of an elaborate integrated set of data allowing adequate individual functional evaluation. In our view, dMRI constitutes an extremely promising imaging modality providing information on the anatomy of white matter tract bundles.

Diffusion tensor imaging is nowadays implemented on many MR systems as a major selling point for manufacturers. However, a large proportion of the neuroscience community remains unaware of the limitations and pitfalls in the current application of this technique. Diffusion tensor imaging and fiber tractography should not be used as a “black box” and conclusions drawn from the data need to be addressed with care and circumspection. Large fiber bundles can be reconstructed as long as they are tagged by an expert along their expected course, and incoherent tracts are identified as spurious fibers and thereby eliminated. Another shortcoming of current tractography algorithms pertains to false negative tracts. As can be appreciated in the corticospinal tract of Figure 13, a significant portion of the usual fiber tract is unrevealed. This limitation of DTI has been explored and extensively reported in recent years. This is explained by the fact that default DTI acquisition commonly uses a 12 direction acquisition averaged three times with a b-value of 1000 s/mm², which is insufficient to reconstruct fiber crossing configurations and obtain robust diffusion measures such as fractional anisotropy, mean diffusivity and apparent diffusion coefficients maps. This limitation is particularly obvious in dense fiber areas where multiple directions intermingle, such as the brainstem, deep

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**Figure 12:** The visual system. The primary visual cortex (Brodmann area 17) was used as the seeding area. Intermingled within these fiber tracts are the inferior fronto-occipital fasciculi.

**Figure 13:** This figure of the corticospinal tract illustrates the limitations of DTI and fiber tractography using current algorithms in complex regions of high curvature fibers as well as fibers crossing, fanning and branching. The red lines indicate several missing and expected fiber tracts according to classical neuro-anatomy.
white matter tracts in close proximity to the thalami and deep basal ganglia, and the corona radiata. Of course, in clinical application and due to overloaded MRI schedules, acquisition time always remains a challenge. Nonetheless, we stress the importance of measuring 60 directions or more diffusion-weighted images to obtain a HARDI (high angular resolution diffusion imaging) dataset from which robust fiber crossing estimation and fiber tractography can be performed.

Although DTI can provide information on white matter tract anatomy, it lacks in terms of functional assessment. This is where functional MRI can complement the diffusion MRI dataset to build a coherent structural/functional network study. We believe that the fusion of fMRI activation and resting-state fMRI maps with current state-of-the-art fiber tractography techniques will allow the study of structural/functional brain networks and that the pre-operative and post-operative evaluation of these networks correlated to the acquisition of neuro-cognitive data will allow a thorough study in the function of the brain, as well as in the role of surgery in glioma patients.

The methodology to perform this fusion of different datasets for each individual patient is currently being developed by our group.

**CONCLUSION**

In this preliminary study, we report state-of-the-art tractography results for some of the major white matter bundles, in a normal subject, using DTI. Moreover, we also used Brodmann’s area as seeding areas for fiber tracts to study the connectivity of known major functional cortical areas. This was accomplished by fusing the 48 Brodmann’s areas into the TI space of our dataset and using the FiberNavigator to select and exclude fiber tracts of interest. This unique integrated dataset was then used to study the visual system in some detail, as well as Wernicke and Broca’s areas, and the primary motor cortex. It is now part of ongoing work to combine HARDI fiber tractography and fMRI activation maps into the FiberNavigator developed by our team to explore functional and anatomical networks in glioma patients.

**REFERENCES**