Determination of the distortion in histological slices of human brain tissue using synchrotron radiation based micro computed tomography

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Optical microscopy is the standard method to characterize the morphology of hard and soft human tissues. Especially the high lateral spatial resolution and the functional staining methods enable us to establish relations between local morphology and function on the cellular level and below. The preparation of histological slices leads to deformations often associated with feature dependent swelling and shrinking, especially for soft matter such as brain tissue. These local distortions are believed to be negligible, because they are just in the percentage range. If the tissue of interest should be represented not only in a 2D image but in a 3D fashion, the histological sections have to be registered, a task, which could become difficult, if the distortions are large. Consequently, it is highly desirable to correct the distortions in the images before their registration in 3D space.

Synchrotron radiation based micro computed tomography (SRμCT) is demonstrated to be an appropriate method to generate 3D data that allow correcting the optical images of the histological brain sections and whereby to determine the local strains in the sections. It has been shown [1, 2] that white and grey matter of human brain tissue exhibit slightly different absorption levels even avoiding any kind of staining. Therefore, the registration of the optical images with the tomographic data provides a reasonable estimate for the local strain as the result of the sectioning procedure.

Human brain tissue was conserved in formalin for several years. For the experiments, parts of the tissue (anterior medulla oblongata including the inferior olivary nucleus) were removed and transferred into Eppendorf containers with a volume of 0.5 mL. To avoid drying artefacts, the containers were entirely filled with phosphate buffer. The container was glued onto the holder and fixed on the high-precision rotation stage of the SRμCT experiment. The measurements were performed at the beamline BW 2 using the standard set-up for absorption contrast tomography. Using the double crystal monochromator, the photon energy was selected to 10 keV, where the absorption difference between white and grey matter could be clearly detected. 721 projections of 1536 x 1024 pixels were recorded. Using an optical magnification of 2.22, the pixel length corresponds to 4 μm. The spatial resolution of 6.2 μm was quantified by the use of the modulated transfer function [3].

Subsequent to the SRμCT experiments the tissue was frozen for crygenic cutting. The 30 μm thick slices underwent optical microscopy in bright- and dark-field mode. Depending on the stain applied, the typical features were visualized (see Figure 1) and registered with the tomographic data. Figure 1 shows an example of a bright-field image of a Nissl stained slice. The nuclei, which are also well distinguished in the SRμCT slices [1, 2], are the conglomerated dark dots. The 2D optical images of the histological slices are elastically registered with the corresponding 2D slices of the tomogram using the multi-modal non-rigid registration algorithm presented in [4]. The result of the registration is depicted by the red-coloured arrows which indicate the direction and amplitude of the local strain.

The combination of the corrected optical 2D images and the 3D SRμCT data allows generating a highly precise atlas of the brain or its parts, which is of importance for minimally or non-invasive surgical interventions and teaching purposes.

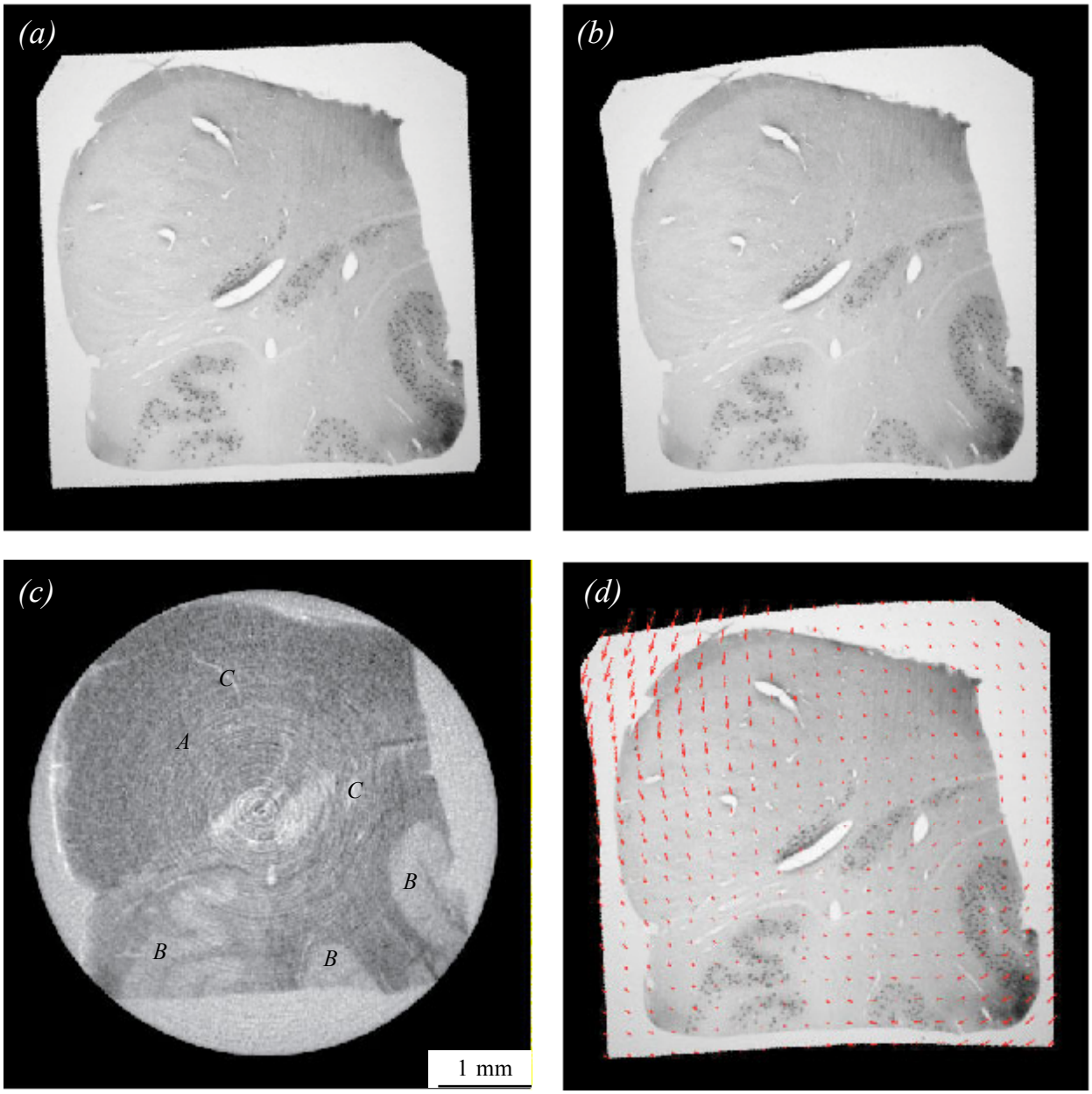

Figure 1: *(a)* The distorted histological slice of brain tissue shows the typical (Nissl-stained) features, i.e. blood vessels as well as the white and grey matter. *(b)* The corrected histological slice can be used, e.g. to rebuild the 3D representation of parts of the brain. *(c)* The letters in the SRμCT slice indicate the white brain tissue – *pyramis* (*A*), the grey matter - *inferior olivary* (*B*), and the blood vessels (*C*). *(d)* The arrows represent the amplitude and direction of the deformation field.

References

[1] M. Germann, F. Beckmann, A. Morel, and B. Müller, HASYLAB Annual Rep. Part I, 1079 (2005)
[2] B. Müller, M. Germann, D. Jeanmonod, and A. Morel, Proc. SPIE 6318, 631803 (2006)
[3] B. Müller et al., Proc. SPIE 4503, 178 (2001)
[4] A. Andronache, Multi-Modal Non-Rigid Registration of Volumetric Medical Images, Ph.D. thesis, ETH-Zurich (2006)